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Clay Modeling in a Sophomore-Level Anatomy Laboratory: Will Active Learning Improve Student Performance?

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Abstract
The purpose of this study was to examine the impact of active learning on student performance in a sophomore-level anatomy and physiology course. Exam grades of students from two consecutive fall semesters were compared. In the first year of the study, students (n=180) used skeletons, plastic muscular manikins, and illustrations to learn the musculoskeletal system while in the second year of the study, students (n=186) also constructed clay models for more active learning. There was no significant difference in average final grade between years, suggesting no difference in overall student ability. For the two laboratory exams over the musculoskeletal system, students who participated in clay modeling performed lower than students who had only skeletons, manikins, and illustrations, and significantly fewer students earned a grade of C (70%) or better on the exams. Surveyed students found active learning useful for visualizing the muscles but few thought clay modeling improved their exam performance. https://doi.org/10.21692/haps.2019.008

Key words: active learning, anatomy laboratory, clay models, formative assessment, student buy-in

Introduction
There is ample evidence that active learning has a positive effect on student performance (Freeman et al. 2014; Michael 2006). Some forms of active learning, however, have been shown to be more effective than others. Lombardi et al. (2014) observed that students who used plastic models, rather than preserved organ dissections or virtual dissections, scored significantly higher on both initial and follow-up exams despite student perceptions that the organ dissections were of more value. Fancovicova and Prokop (2014) noted that students who had access to multiple forms of active learning performed at a higher level than students who had access to only one form of active learning. Herur et al. (2011) found that students who participated in active learning had higher levels of retention 15 and 30 days after the class than those who learned by passive means. The level of student engagement has also been shown to impact learning. LaDage et al. (2018) found that students who engaged in direct manipulation of a model, as compared to students who merely watched another person manipulate the model or listened to a lecture, performed significantly better on initial assessment. The results of this study, however, did not demonstrate any differences in retention based on learning technique. Kooloo et al. (2014), however, observed that students who attentively observed others build clay models showed higher increases in anatomical knowledge than those students building the models and concluded that engagement in and focus on the task were keys to learning.

Clay modeling has been used to promote understanding of internal organs (Shipley, 2010), the brain (Akle et al. 2018; Kooloos et al. 2014), the nervous system (Herur et al. 2011), and the musculoskeletal system (Bareither et al. 2013; DeHoff et al. 2011; Mutoike et al. 2009; Waters et al. 2005; Water et al. 2011). Mutoike et al. (2009) found that students who built clay models on human manikins were better able to identify muscles on human models than were students who performed cat dissection. Students who constructed clay models also performed higher on exams than students who performed preserved organism dissection (DeHoff et al. 2011; Waters et al. 2005). Bareither et al. (2013) observed that, while students who participated in active learning showed greater increases in knowledge gain than students who did not engage in active learning, there were no differences among students who built clay models compared to students who completed written modules. Moreover, there were no differences in three-month retention between the groups.

Students who are the first in their immediate family to attend college (i.e., first-generation students) have been shown to benefit from active learning (Eddy and Hogan 2014), and generation status has been shown to be more influential than other fixed characteristics on student buy-in to active learning (Brazeal and Couch 2017). At the University of Nebraska at Kearney, first-generation students make up better than 40% of undergraduate enrollment. Therefore, when funds became available over the summer, new supplies in the form...
of clay models were purchased with the goal of improving student learning through active learning techniques. In previous years students learned muscle attachments and actions using information provided in tables, illustrations, isolated bones, skeletons, and muscular manikins only. It was anticipated that creating the clay models, where students could place the muscles on the models and better visualize the attachments and actions, would improve student learning. The purpose of this study was to determine if supplementing the current laboratory teaching materials with active learning through the construction of clay models would improve student performance on exams over the anatomy of the musculoskeletal system in a sophomore-level anatomy and physiology course.

**Methods**

Approval was obtained from the Institutional Review Board at the University of Nebraska at Kearney (protocol 010919-1; exempt status). The study used existing data from students enrolled in Biology 225, Anatomy and Physiology I, Fall 2017 and Fall 2018 semesters. Biology 225 is the first of a two-semester course sequence and college-level chemistry is the required prerequisite. In addition, students enrolled in Biology 226 Anatomy and Physiology II spring 2019 were surveyed regarding their opinions of the utility of the clay models they had used in laboratory the previous semester.

There were nine laboratory sections both semesters, each with one instructor and as many as 24 students. Plastic human half-skeletons and clay kits (Anatomy in Clay®, Zahourek Systems, Inc. and Affiliates, Loveland, CO) were purchased in the summer of 2018. Enough kits were purchased so that if every lab was at maximum enrollment students could still work in pairs. The investment per student in purchasing these kits was $162, approximately eight times the $20 laboratory fee for the course.

The laboratory portion of the course was divided into three, five-week sections. Each section included four weeks of laboratory activities followed by a laboratory practical exam. The first laboratory practical exam covered basic terminology and organization, movements through membranes, tissues, integumentary system, basic microscopic and macroscopic anatomy of bones, and types of joints. The second laboratory exam covered the bones and bone features, joints, and muscles of the upper body plus neuromuscular physiology. Students constructed clay models for three of the four laboratory periods prior to the exam. The third laboratory exam covered the bones and bone features, joints, and muscles of the lower body plus central nervous system anatomy. Students again constructed clay models for three of the four laboratory periods prior to the exam. Weekly laboratory activities were identical each year and each week's laboratory had clear objectives. The laboratory was not dissection or cadaver-based. Students used complete skeletons (e.g., Max the Classic Skeleton with Muscle Insertions and Origins, catalog number S-A11, 3B Scientific, Tucker, GA), isolated bones, muscular manikins (e.g., ¾ Life-Size Dual-Sex Muscle Figure, 45-part, catalog number S-B50, 3B Scientific, Tucker, GA), isolated limb muscular models (e.g., ¾ Life-Size Muscle Arm, 6-part, catalog number S-M10, 3B Scientific, Tucker, GA), and illustrations, to learn the required structures and were tested using these same materials. Students in Fall 2018 also worked in pairs to create clay models of the musculature on human half-skeletons, which were used for the laboratories covered in the second and third laboratory exams. Following each laboratory introduction that highlighted the regional bones, muscles, and joints, the students were allowed to work with the models and/or complete other laboratory exercises. The students were instructed to pay attention to certain features of the muscles that would be important when constructing the models. These features included whether the muscles were superficial or deep as well as their origins and insertions. Students were not told the order of muscles to place on the models or how many they should complete that week. No instructions were given on the amount of clay or the amount of detail that should be used in each muscle design. Students were encouraged to use the models as a learning tool and knew they were not going to be graded on the activity. Completed student-built clay models were used in laboratory examinations two and three in 2018 (Fall semester).

All laboratory exams required students to observe physical displays and answer questions about the display. Each exam had 25 stations with either three questions (exam one; 75 points possible) or four questions (exams two and three; 100 points possible) per station, and students had one hour and 45 minutes to complete the exam. Students were permitted to go to the stations in any order and could return to a station as many times as they wanted to during the exam period. Students had an alphabetical list of potential terms they could use to answer the questions and credit was deducted for spelling errors. The terms on the list had to be combined for multiple-word answers, and students wrote their answers in numbered blanks on a paper answer sheet. Examples of typical questions for exams two and three from a muscular manikin, an illustration, a clay model, and an isolated bone are shown in Figure 1.
Clay Modeling in a Sophomore-Level Anatomy Laboratory: Will Active Learning Improve Student Performance?

The questions and set up of the exams were identical with the exception of substituting student-built clay models as the visual display for about 10 questions on exams two and three in 2018 (Fall semester).

With the exception of the clay modeling activity and university closure on a day in which lab exams were given that resulted in rescheduling, all other aspects of the course, including the content, number, and scheduling of lecture exams as well as the timing of Fall break and Thanksgiving break relative to laboratory exams, were identical each year. Only students who completed all aspects of the course were included in data analysis. Exam performance (percentage correct) was analyzed between years using a student’s two-tailed t-test. Student performance within year and between years was analyzed using a two-way ANOVA. Chi-squared analysis was performed to determine if there were a difference in the percentage of students in the classes who earned a grade of C (70%). Significance was ascribed for p<0.05.

Results

A total of 180 and 186 students completed all aspects of the course in Fall 2017 and Fall 2018, respectively. There was no difference in student performance on exams one or two between years, but student performance on exam three was significantly higher (p = 0.004) in 2017 (Table 1).
In 2017 there was a significant difference in student performance on the exams within year but in 2018 there was no difference in student performance within year (F (2,728) = 17.42, p < 0.0001). Chi-squared analysis determined there was no difference in the percentage of students earning a final grade of C or higher between years but there was a significant difference in the percentage of C or higher grades on the practical exams (Table 2).

A total of 50 students out of the 150 students enrolled in Biology 226 completed the survey (Table 3). In general, a majority of students agreed that the clay models improved their ability to visualize layering of the muscles and muscle attachments, but opinions were more evenly split regarding whether the clay models helped them see muscle agonists and antagonists or were a valuable learning tool. Most students disagreed that the clay models improved their exam performance.

<table>
<thead>
<tr>
<th>Year</th>
<th>Exam 1</th>
<th>Exam 2</th>
<th>Exam 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>2017 (n=180)</td>
<td>77.2 ± 13.2</td>
<td>79.8 ± 13.6</td>
<td>82.1 ± 16.8</td>
</tr>
<tr>
<td>2018 (n=186)</td>
<td>78.4 ± 14.1</td>
<td>76.7 ± 17.2</td>
<td>76.2 ± 21.4</td>
</tr>
</tbody>
</table>

* significantly different from exam three, Fall 2017, p = 0.004

**Table 1.** Laboratory practical exam results (average percentage correct ± SD) for Anatomy and Physiology I students who completed all aspects of the course. Exam questions were identical both years. In 2018 laboratory exercises covering the musculoskeletal system in preparation for exams two and three were supplemented with clay modeling activities, and these models were used for some exam questions. Data were analyzed using a student’s two-tailed t-test and two-way ANOVA. Significance was ascribed for p<0.05.

<table>
<thead>
<tr>
<th>Year</th>
<th>Exams 2 and 3</th>
<th>Final grade</th>
</tr>
</thead>
<tbody>
<tr>
<td>2017</td>
<td>81.4</td>
<td>79.4</td>
</tr>
<tr>
<td>2018</td>
<td>72.3</td>
<td>76.9</td>
</tr>
</tbody>
</table>

**Table 2.** Percentage of students earning a grade of C or better (70% or higher). Chi-squared analysis determined that the difference in the percentage of C or better grades on exams two and three was significantly different between years. The percentage of students earning a final grade of C in the class, however, was not significantly different between years.

<table>
<thead>
<tr>
<th>Statement</th>
<th>Agree</th>
<th>Disagree</th>
<th>Neutral</th>
</tr>
</thead>
<tbody>
<tr>
<td>The clay models improved my ability to visualize the layering of the muscles.</td>
<td>74</td>
<td>20</td>
<td>6</td>
</tr>
<tr>
<td>The clay models improved my ability to visualize the muscle attachments.</td>
<td>54</td>
<td>40</td>
<td>6</td>
</tr>
<tr>
<td>The clay models allowed me to better visualize antagonistic muscle groups.</td>
<td>48</td>
<td>40</td>
<td>12</td>
</tr>
<tr>
<td>The clay models were a valuable learning tool.</td>
<td>46</td>
<td>48</td>
<td>6</td>
</tr>
<tr>
<td>The clay models allowed me to make connections between muscles with similar actions.</td>
<td>44</td>
<td>42</td>
<td>14</td>
</tr>
<tr>
<td>The clay models improved my performance on the laboratory exams.</td>
<td>30</td>
<td>54</td>
<td>16</td>
</tr>
</tbody>
</table>

**Table 3.** Results of survey asking students the utility of clay models in the anatomy and physiology laboratory. A total of 50 students completed the survey. Numbers represent percent of responses.
Discussion
The results of this study suggest that inclusion of the hands-on modeling activity did not increase student learning as measured by exam performance; in fact, student performance was lower than the previous year on exam two (p=0.063) and significantly lower than the previous year on exam three. It is important to note that student performance on laboratory exam one serves as a control in that the course materials were presented in identical fashion both years and there was no difference in student performance on that exam. In addition, the final overall grades for students were also not significantly different (2017 average 78.8 ± 11.7 percent; 2018 average 77.7 ± 13.6 percent). Therefore, the overall academic performance of students participating in the study each year was similar. Moreover, four of the five laboratory instructors were the same both years, the laboratory exercises were organized in such a way that learning objectives were clear, and grading instructions and criteria for exams were very clear so as to ensure consistency in grading across instructors. Therefore, it is not unreasonable to suggest that differences in student performance on exams two and three were influenced by the learning materials.

Student performance on exam one was the lowest of the three exams in 2017 and student performance increased on each subsequent exam. In contrast, the highest average of the three exams in 2018 was exam one and student performance slightly, but not significantly, decreased with each subsequent exam. During the previous ten years (2007-2016), student performance in this class was lowest on the first practical exam six times and highest on the second practical exam nine times; student performance on the third exam was lowest three times and highest once. 2018 was the only time during this 12-year period (2007-2018) that student performance on the practical exams decreased with each subsequent exam.

Student interest in the clay models waned noticeably over the course of the semester to the extent that many students did not use the clay models at all in preparation for exam three. This is consistent with the results of our survey (Table 3) in which the majority of students indicated they did not think clay modeling improved their performance on the exam. As observed by Brazeal and Couch (2017), students must buy in to the approach in order for it to have an impact on learning. If the learning activity promotes activity for the sake of activity but not student learning, student disengagement will result (Smith and Cardaciotto 2011). In addition, the amount of structure provided by the instructor has an impact on exam performance (Reinhardt and Rosen, 2012). In order to keep the laboratory experiences similar, no additional direction or structure was provided to the students in the current study as to how to incorporate the clay models into their learning. There was an example model to which laboratory instructors built a muscle each laboratory period, but this was the only additional direction provided.

It is critical that faculty employ proven methods to incorporate effective active learning into their class (Goodman et al. 2018). Clay models have been shown to be an effective learning tool (Akle et al. 2018; DeHoff et al. 2011; Haspel et al. 2014; Oh et al. 2009; Waters et al. 2005; Waters et al. 2011), but it has also been shown that students and faculty have different perceptions regarding active learning (Tsang and Harris 2016). No course credit was assigned to the clay models, so even though students worked in pairs some individuals were left to do all the work while their partners did little or nothing. This led to frustration and questions from students as to whether or not their non-participatory partner could lose credit in some way for not helping create the clay model. Some students, including very high achieving students, focused only on the immediate value of the activity in terms of course credit (or lack thereof) in the form of points.

Adding course credit (points) to the clay modeling activity would most likely improve participation but, given that the majority of students did not think clay modeling improved their exam performance (Table 3), activity without a perceived purpose is not an effective learning strategy (Smith and Cardaciotto 2011). Brazeal and Couch (2017) demonstrated that unfixed student qualities, such as their perception of whether or not the activity is relevant or challenging, were a higher predictor of student buy-in than fixed student qualities. Akle et al. (2018) observed that some students viewed clay modeling as juvenile at first but came to realize the activity contributed in a meaningful way to the learning process.

Disinterest by some high-achieving students may have led marginally-performing students who might have benefited from the hands-on activity to abandon the models. The significant decrease in the percentage of students earning a grade of C or better on the exams may reflect not only abandonment of the clay models but failure to then follow through on the existing formative assessment techniques for the skeletons, manikins, and models. While faculty are often reluctant to try new techniques due to the time commitment to develop the technique (Miller and Metz 2014) or anticipated student resistance (Brazeal and Couch 2017; Smith and Cardaciotto 2011), the instructors in the current study were excited to have a new hands-on learning technique and the lack of student buy-in was not expected.

Fancovicova and Prokop (2014) found that students who used a combination of active learning methods showed greater achievement than students who used only one active learning method. While we had hoped the addition of clay modeling would have improved student performance, we already used multiple teaching tools in our laboratory; therefore, the addition of the clay modeling could have been one modality too many for some students and overwhelmed them. As Kooloos et al. (2014) concluded, students must focus their attention and engage in the exercise in order for learning to take place. Our laboratory period was one hour and 50 minutes, which included
time for a weekly quiz, direction from the instructor, getting supplies ready, and cleaning up the workspace. Therefore, students often had only 60-75 minutes to work on the clay models, and it was challenging for some students to complete the clay models during the laboratory period. Each week an average of eleven new muscles were assigned (range eight to fourteen), so students had as little as five minutes to devote to each new muscle and may have hurried to build the models without focusing on what it was they were to learn. That left little if any time for the students to make use of the other learning materials available to them, such as the muscular manikin and isolated muscular limb models. In order to make the clay modeling more effective in future, the labs might be redesigned to have fewer muscles each week with muscles being included in more labs throughout the semester as well as clear objectives with the clay model outlined in the student’s manual. Alternatively, fewer muscles could be assigned to the clay models with specific questions assigned. For example, in the laboratory exercise focusing on muscles of the hip and thigh, perhaps only the muscles of the quadriceps group might be assigned to the clay model with specific questions regarding the order in which they must be placed on the model. This could help reinforce concepts related to “superficial” versus “deep” muscles as well as reinforce why the rectus femoris has different actions from the other muscles of the quadriceps group.

In order to keep the classes as identical as possible so as to assess the impact of the clay models themselves, no changes were made to the instructions for the laboratory activities or to the in-class formative assessment worksheets. The lack of any formative assessment techniques specific to the clay models, however, likely had a role in both student buy-in and subsequent exam performance. Haspel et al. (2014) successfully incorporated these same clay models in the anatomy and physiology laboratory at a large community college. Incorporation of the clay models into the laboratories at the community college was not done, however, until the instructors had undergone specialized training in their use, a new custom laboratory manual had been developed, and all the instructional materials had been completely revised. With this extensive preparation, 96% of the faculty felt the clay modeling was an effective learning experience. Student performance on exams improved significantly over the previous year, but at the end of the term only 51% of the students felt clay modeling was a positive experience (Haspel et al. 2014).

At our institution, the clay models had been on a “wish list” for years, were eagerly purchased when funds became available, and were incorporated into the classroom scarcely a month later as a supplement to the existing materials. In conclusion, the results of this study suggest that active learning strategies must be employed with proper considerations to factors such as student involvement, instructional details, and time restrictions of the laboratory period in order to see improved student performance.

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Janet Steele, PhD, is a Professor of Biology at UNK. Her primary teaching responsibilities are undergraduate anatomy and physiology lecture and laboratory.

Literature cited


Clay Modeling in a Sophomore-Level Anatomy Laboratory: Will Active Learning Improve Student Performance?


Teaching Assistants in Human Anatomy and Physiology: Their Prevalence, Recruitment Strategies, Funding, Retention, and Training

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Abstract
Studies show teaching assistants (TAs) can positively influence the learning environment by increasing student comprehension and retention and impacting students’ choices of academic and career paths. However, use of TAs in higher education is not universal. Following a fruitful panel discussion at the 2017 HAPS Conference, we conducted a survey to assess the prevalence of undergraduate and graduate TAs in human anatomy and physiology courses across a range of institutions and to evaluate how TAs are recruited, trained and compensated. Data from 329 respondents who used TAs, indicate most institutions use a formal application process, training procedures vary, and most TAs are compensated financially through hourly wages, stipends, or tuition credit. However, some TAs are unpaid but receive course credit as compensation. We highlight one of the more successful recruiting and training programs, where paid master TAs mentor unpaid apprentice TAs. This tiered training program excites and motivates student leadership, maintains high academic standards, and alleviates the lead instructor from continually training novice TAs. https://doi.org/10.21692/haps.2019.015

Key words: teaching assistant, anatomy, physiology, laboratory, education

Introduction
Teaching assistants (TAs) have, of necessity, been part of science laboratory course instruction for more than four decades in North America. For example, use of TAs offers many benefits including freeing up faculty time for research; staffing teaching labs and recitations associated with large enrollment undergraduate lecture courses; and providing teaching apprenticeship experiences along with financial support for student TAs (Park 2004). TAs gain methodological research skills in the process (Feldon et al. 2011, Schalk et al. 2009). Having additional instructional personnel also facilitates implementation of active learning, particularly inquiry-based laboratory lessons (Hughes and Ellefson 2013). More studies focus on graduate TAs rather than undergraduate TAs. However, it is known that peer-to-peer instruction positively impacts student learning by removing student hesitation to approach the lead instructor (Wheeler et al. 2017) and improves student retention in STEM fields (O’Neal et al. 2007).

The purpose of this study was to assess the extent to which lead Human Anatomy and Physiology instructors utilize TAs, and if their experiences reflect the benefits reported in previous studies for general science courses. Because Anatomy and Physiology is taught at a variety of institutions from small two-year community colleges to large doctoral degree granting research institutions, we sought to develop a comprehensive picture about how TAs in Anatomy and Physiology were being trained, compensated, utilized (lecture, lab, or online; teaching or grading), and their perceived impact. Training is paramount, especially for undergraduate TAs, as their role shifts from learner to a guide, mentor, and facilitator. The content load of an Anatomy and Physiology course is extensive and training graduate TAs who have not previously taken the course is time consuming and reduces the amount of time available for teaching students enrolled in the courses. With this nationwide study we hope to identify unifying training components like Family Educational Rights and Privacy Act (FERPA), lab safety, and pedagogy, and provide insight about concerns lead instructors have about their own time commitment to the process, which in some cases is neither compensated nor considered part of their teaching load.

The authors have had a combined experience of 120 years of supervising TAs and wanted to share their insight on the best practices for building a successful TA program. This study may initiate establishing guidelines regarding criteria for supervising and utilizing TAs as well as future scholarship of teaching and learning projects, both of which will ultimately influence student success.
Materials and Methods
After a successful panel discussion on Effective Recruitment, Retention and Training for Teaching Assistants in Anatomy and Physiology at the HAPS 2017 conference, the authors designed a survey to assess TA use in Human Anatomy and Physiology courses. The survey was developed and administered via Qualtrics survey software with the text and flow sequence of survey questions listed in Appendix A. Subjects were recruited through email announcements distributed via the Human Anatomy and Physiology Society (HAPS) listserv from 24 October 2017 to 15 November 2017. Upon completion of the anonymous survey, respondents were given the option to enter a raffle for one of five Amazon gift cards funded by HAPS, one for $100 and four for $50, in appreciation for completing the survey. Winners of the raffle were randomly selected on 16 November 2017 and gift cards were distributed in January 2018. This study (Protocol #18x-061) was reviewed by The University of Mississippi’s Institutional Review Board (IRB) and was approved as Exempt under 45 CFR 46.101(b) (#2).

Survey responses were coded by institution type, e.g., 2-year community/technical college; 4-year private, non-profit; 4-year public, non-profit; for profit; and other, prior to statistical analyses. Numerical and text responses to select questions from 338 respondents are presented in the areas of: respondent profiles; reasons for using TAs; reasons for not using TAs; selection and training of TAs; compensation for TAs; benefits and drawbacks to TAs in the laboratory classroom; and future issues regarding TA involvement in Human Anatomy and Physiology courses. Where respondents could enter a numerical response rather than select a categorical response, descriptive statistics such as mean and standard error were calculated to inform the reader of variation across the survey responses.

Results
Survey participants
A total of 338 educators participated in our survey. Out of the 338 respondents, nine participants were excluded from the rest of the study because they reported that colleagues used TAs, but they themselves did not, and therefore were not able to answer subsequent questions about how TAs are selected, trained, utilized, and compensated. Responses were received mostly from the United States, but there were a few (<10) from Canada, Africa, and Australia. Survey results were compiled based on public vs. private institutions as well as four-year vs. two-year colleges and community colleges. Most participants taught either at a two-year community/technical school (41.34%) or a four-year public, nonprofit institution (36.78%; Table 1). Individuals who identified as teaching in the “other” category provided responses that included government, graduate programs or graduate allied health programs, as well as public high schools, and branch campuses of larger institutions.

<table>
<thead>
<tr>
<th>Institution</th>
<th>Participant number (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2-year community/technical</td>
<td>136 (41.34%)</td>
</tr>
<tr>
<td>4-year private, nonprofit</td>
<td>58 (17.63%)</td>
</tr>
<tr>
<td>4-year public, nonprofit</td>
<td>121 (36.78%)</td>
</tr>
<tr>
<td>For profit</td>
<td>3 (0.91%)</td>
</tr>
<tr>
<td>Other</td>
<td>11 (3.34%)</td>
</tr>
<tr>
<td>Total</td>
<td>329 (100%)</td>
</tr>
</tbody>
</table>

Table 1. Number of survey participants by institution type.

All institutions reported offering less than 20 Anatomy and Physiology lecture sections during a typical semester with the number of students per lecture varying by institution type (Figure 1). In general, the number of Anatomy and Physiology lectures ranged from five sections in private institutions to 19 sections in two-year community/technical schools. Four-year non-profit institutions had the highest mean number of students per lecture (170), while two-year community/technical schools, which offered the largest number of Anatomy and Physiology sections, had a mean of 37 students per class (Figure 1). The number of Anatomy and Physiology lab sections did not differ as much by institution. Both four-year public, nonprofit and two-year community colleges offered an average of 22 to 24 lab sections, ranging between 37-42 students per section. Four-year private, nonprofit institutions had approximately 13 sections with 46 students per section on average, and for-profit institutions have up to six lab sections. Schools that classified themselves in the “other category” did not report teaching Anatomy and Physiology labs (Figure 2).
Figure 1. Respondent profile of Human Anatomy and Physiology lecture offerings, student enrollment in offered sections, and number of years using TAs. Columns represent the mean numerical response across institution type with error bars representing ± 1 standard error around the mean.

Figure 2. Respondent profile of Human Anatomy and Physiology lab offerings, student enrollment in offered sections, historical use of teaching assistants, and teaching assistant to student ratios. Columns represent the mean numerical response across institution type with error bars representing ± 1 standard error around the mean.
Use of Teaching Assistants

All institutions, except for-profit institutions, reported using TAs in their Anatomy and Physiology courses in some capacity (Figure 1). Instructors that chose not to use TAs reported budget and funding limitations as their main reasons in the two-year community/technical schools, followed by fill-in responses that ranged from administrative expectations that only faculty or graduate students (in the four-year public, nonprofit institutions) teach, and lack of necessity for given class sizes (Figure 3). Other reasons for not utilizing TAs included lack of desire, concerns about having undergraduate students have access to grades, and perceived lack of TA quality.

Figure 3. Survey respondents who said they did not use TAs were directed to questions regarding their reasons for not using TAs. Respondents could select multiple reasons. The “Blank” respondents use TAs and skipped this question. Four-yr public, nonprofit and four-yr private, nonprofit are the leading users of TAs per this survey question. For two-yr community/technical college respondents, budget/funding was a chief reason for not using TAs. The write-in responses (other) for two-yr college respondents indicated that class sizes were small and only the faculty were expected to teach, high turnover or TAs expected, and comments alluding to budget limitations.
The TAs in four-year institutions are generally a mix of undergraduate and graduate students, primarily graduate students. In the two-year community/technical schools, TAs are mainly undergraduate students and rarely adjuncts (Table 2).

Teaching assistants are utilized in different capacities and diverse classrooms. For example, TAs are primarily used in four-year public, non-profit institutions, to lead group discussion and to help students during active learning activities. They are used mostly in the labs and sometimes in the lecture class, as well as offering open labs and helping professors with preparation. To a much lesser extent, TAs are utilized in hybrid and online courses by two-year community/technical, and the four-year institutions (Table 2). In four-year institutions (Table 2), TAs typically teach content either as the sole instructors in the lab room, primarily in four-year public nonprofit institutions, or alongside other instructors or TAs, in both public and private four-year institutions.

<table>
<thead>
<tr>
<th>Classification of teaching assistants</th>
<th>Categorical response</th>
<th>2-yr community/technical</th>
<th>4-yr private, nonprofit other</th>
<th>4-yr public, nonprofit</th>
<th>for profit</th>
<th>other</th>
</tr>
</thead>
<tbody>
<tr>
<td>Both undergraduate and graduate students</td>
<td>0</td>
<td>8</td>
<td>33</td>
<td>0</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Graduate students</td>
<td>1</td>
<td>2</td>
<td>27</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Undergraduate students</td>
<td>10</td>
<td>20</td>
<td>16</td>
<td>0</td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>Adjunct</td>
<td>3</td>
<td>5</td>
<td>9</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Teaching assistant autonomy</th>
<th>Classification</th>
<th>Categorical response</th>
<th>2-yr community/technical</th>
<th>4-yr private, nonprofit other</th>
<th>4-yr public, nonprofit</th>
<th>for profit</th>
<th>other</th>
</tr>
</thead>
<tbody>
<tr>
<td>TAs teach without an instructor in the room</td>
<td>3</td>
<td>5</td>
<td>46</td>
<td>0</td>
<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>TAs assist alongside lab instructors (or additional TAs) who are always in the room</td>
<td>1</td>
<td>1</td>
<td>7</td>
<td>0</td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td>2</td>
<td>2</td>
<td>5</td>
<td>0</td>
<td>0</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Course format</th>
<th>Categorical response</th>
<th>2-yr community/technical</th>
<th>4-yr private, nonprofit other</th>
<th>4-yr public, nonprofit</th>
<th>for profit</th>
<th>other</th>
</tr>
</thead>
<tbody>
<tr>
<td>Face-to-face lab</td>
<td>6</td>
<td>26</td>
<td>73</td>
<td>0</td>
<td>3</td>
<td></td>
</tr>
<tr>
<td>Face-to-face lecture</td>
<td>5</td>
<td>8</td>
<td>30</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Hybrid lecture</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Online lecture</td>
<td>2</td>
<td>0</td>
<td>3</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td>2</td>
<td>0</td>
<td>3</td>
<td>0</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Hybrid lab</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Online lab</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>

Table 2. Classification and autonomy of teaching assistants in anatomy and physiology classes, and course format, by institution type. Respondents were able to select more than one answer. Categorical responses are ranked by number of responses (from high to low) at four-year public, nonprofit institutions (i.e., the institution category with the highest response rate to these questions on the survey).
In addition to active laboratory instruction, TAs participate in a range of course activities, during class time and outside of class (Table 3). During class, TAs help the instructor address student questions and guide students through active learning exercises. They participate in station work, proctor exams, and run errands. Outside of class, TAs offer tutoring and host open labs, as well as setup and breakdown labs, and prepare reagents (Table 3).

<table>
<thead>
<tr>
<th>Student interactions</th>
<th>Categorical responses</th>
<th>2-yr community/technical</th>
<th>4-yr private, nonprofit other</th>
<th>4-yr public, nonprofit</th>
<th>for profit</th>
<th>other</th>
</tr>
</thead>
<tbody>
<tr>
<td>Station work</td>
<td></td>
<td>7</td>
<td>20</td>
<td>50</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Staff open lab times</td>
<td></td>
<td>6</td>
<td>20</td>
<td>46</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Help the lead lab instructor during the lab whenever students need help</td>
<td></td>
<td>6</td>
<td>26</td>
<td>41</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Conduct practice lab practicals</td>
<td></td>
<td>0</td>
<td>1</td>
<td>36</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Other</td>
<td></td>
<td>1</td>
<td>1</td>
<td>27</td>
<td>0</td>
<td>1</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Additional responsibilities</th>
<th>Categorical responses</th>
<th>2-yr community/technical</th>
<th>4-yr private, nonprofit other</th>
<th>4-yr public, nonprofit</th>
<th>for profit</th>
<th>other</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clean up lab</td>
<td></td>
<td>4</td>
<td>25</td>
<td>46</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Set up lab</td>
<td></td>
<td>3</td>
<td>26</td>
<td>46</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Guide students through active learning exercises</td>
<td></td>
<td>6</td>
<td>12</td>
<td>42</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Running errands</td>
<td></td>
<td>2</td>
<td>9</td>
<td>18</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Prepare chemical reagents</td>
<td></td>
<td>1</td>
<td>14</td>
<td>15</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Other</td>
<td></td>
<td>4</td>
<td>2</td>
<td>9</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>None of these are done by my TAs</td>
<td></td>
<td>2</td>
<td>1</td>
<td>9</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Table 3. Student interaction opportunities and additional responsibilities of teaching assistants in anatomy and physiology classes by institution type. Categorical responses are ranked by number of responses (from high to low) at four-year public, nonprofit institutions (i.e., the institution category with the highest response rate to these questions on the survey). Respondents were able to select more than one answer.
Due to institutional policies, graduate TAs have the greatest latitude in grading and undergraduate TAs the least. Grading is primarily observed in the four-year institutions, where TAs may write and grade lab quizzes and practicals, as well as enter the grades into the learning management system (Figure 4). Out of all participants, approximately 25% report having TAs actively involved in creating assessments (quizzes, practicals, exams) and 45% report having TAs participate in some form of grading. Teaching assistants grade primarily multiple choice, short answer, and labeling questions (Figure 5). 7.73% of participants do not assign grading responsibilities to their TAs. In addition to formal grading, 5.5% of participants report having their TAs grade assignments and check homework or lab reports. The decision to allow TAs to grade appears dependent on institutional policies, and undergraduate TAs at many institutions are not allowed to participate in any form of grading.

Figure 4. Grading responsibilities of teaching assistants in anatomy and physiology laboratory classrooms. Respondents were able to select more than one answer. Where an institution category appears in the figure legend but does not show in the stacked columns, there were no selections from respondents at that type of institution for this question.

Figure 5. Question types graded by teaching assistants in anatomy and physiology laboratory classrooms. Respondents were able to select more than one answer. Where an institution category appears in the figure legend but does not show in the stacked columns, there were no selections from respondents at that type of institution for this question.
TA selection, training, and compensation

Teaching assistants are typically selected based on multiple criteria. All respondents that use TAs report criteria of a formal application, academic success, and/or observations of the applicant in an Anatomy and Physiology course while a student, as well as interviews and references (Table 4). In addition, participants who chose “Other” reported hiring decisions based on completion of a Bachelor’s degree (or teaching internship) or appointment decisions made by graduate program directors, TA coordinators, or graduate committees. Selection of graduate TAs seems to be based more on the need for the TAs to acquire teaching experience as part of their graduate education than their background in human anatomy and physiology.

<table>
<thead>
<tr>
<th></th>
<th>2-yr community/technical</th>
<th>4-yr private, nonprofit other</th>
<th>4-yr public, nonprofit</th>
<th>for profit</th>
<th>other</th>
</tr>
</thead>
<tbody>
<tr>
<td>Application</td>
<td>5</td>
<td>10</td>
<td>40</td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>High grade in the course</td>
<td>7</td>
<td>18</td>
<td>39</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Observations of positive behavior by the student when they were taking the course</td>
<td>8</td>
<td>22</td>
<td>33</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Interview</td>
<td>4</td>
<td>7</td>
<td>24</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Other</td>
<td>3</td>
<td>7</td>
<td>24</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>References</td>
<td>5</td>
<td>6</td>
<td>20</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Table 4. Selection and hiring procedures for teaching assistants in anatomy and physiology classes by institution type. Responses are ranked by number of responses (from high to low) at four-year public, nonprofit institutions (i.e., the institution category with the highest response rate to this question on the survey). Respondents were able to select more than one answer.
Once hired, 65% of TAs receive training either directly from the instructor (44%) or via a formal training course (21%). 24.6% of TAs do not receive any training, or training is conducted through the institution, by more senior TAs, or a TA coordinator (10%), (Table 5). The three main topics covered through formal training include FERPA, lab safety, and pedagogy (Table 5). Other topics include Title IX training, instruction on how to help students learn, and observations of teaching labs. Training can also occur through TA meetings held with variable regularity among institutions.

<table>
<thead>
<tr>
<th>Categorical response</th>
<th>2-yr community/technical</th>
<th>4-yr private, nonprofit other</th>
<th>4-yr public, nonprofit</th>
<th>for profit</th>
<th>other</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yes, I train them myself (no course for TAs)</td>
<td>3</td>
<td>13</td>
<td>35</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Yes, there is a formal course for training TAs</td>
<td>5</td>
<td>2</td>
<td>17</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>No</td>
<td>1</td>
<td>12</td>
<td>14</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Other</td>
<td>0</td>
<td>2</td>
<td>10</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Lab Safety</td>
<td>7</td>
<td>13</td>
<td>51</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Pedagogy</td>
<td>6</td>
<td>8</td>
<td>41</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>FERPA student data privacy issues</td>
<td>4</td>
<td>8</td>
<td>37</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Mentors and apprentices</td>
<td>3</td>
<td>6</td>
<td>32</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Other</td>
<td>1</td>
<td>1</td>
<td>6</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Academic credit training course</td>
<td>2</td>
<td>2</td>
<td>3</td>
<td>0</td>
<td>1</td>
</tr>
</tbody>
</table>

Table 5. Components of formal training programs and training procedures for teaching assistant compensation in anatomy and physiology classes by institution type. Categorical responses are ranked by number of responses (from high to low) at four-year public, nonprofit institutions (i.e., the institution category with the highest response rate to these questions on the survey).
Compensation
Compensation for TA (Figure 6). In some cases, undergraduate TAs might receive course credit for being a TA (Figure 7), but that seems to be very rare.

Figure 6. Financial compensation for teaching assistants in anatomy and physiology classes. Where an institution category appears in the figure legend but does not show in the stacked columns, there were no selections from respondents at that type of institution for this question.

Figure 7. Course credit-based compensation for teaching assistants in anatomy and physiology classes. Where an institution category appears in the figure legend but does not show in the stacked columns, there were no selections from respondents at that type of institution for this question.
**Discussion**

The aim of this study was to assess the prevalence and extent of TA use in Human Anatomy and Physiology courses. Thirty-nine percent of our participants use TAs in their Anatomy and Physiology courses. Of the 149 participants (59%) that do not utilize TAs, but also clarified that they ‘would like to do so’ or ‘do not wish to have TAs’, 76 reported wanting to be able to use TAs. For many participants in this latter category, their inability to use TAs was mainly due to budgetary constraints and lack of administrative support. Where TAs are part of the course structure, the TA composition, responsibilities, training and compensation vary, presenting multiple opportunities and challenges to incorporation of TAs into the classroom. The authors of this paper assert that TAs add value to Anatomy and Physiology instruction and offer solutions to the presumed barriers to implementation.

**Value of TAs to Instruction**

Teaching assistants contribute in multiple ways to course organization and instruction. Our survey showed that instructors utilize TAs outside of class to help prepare materials for labs and clean up after lab, grade various aspects of assessments, help tutor, and hold open labs. Being able to rely on responsible, well-trained TAs to help manage a course, especially in institutions with high enrollment, can improve the types of learning experiences the instructors can offer. For example, where TAs participate in course instruction, it is possible to divide a large class into smaller groups using a station-based approach (Goldina and Barattini 2019; Springer et al. 1999, Whelan et al. 2016), which is used by many instructors in four-year institutions.

In the classroom, TAs can be extremely useful in helping students receive individualized attention and decreasing the length of time students must wait for assistance. In institutions where TAs were used, regardless of institution type, instructors noted that TAs spent most of their time engaging with students (Table 6), which can be beneficial on many levels. Giving students the opportunity to interact with TAs, who are often much closer in age to the students, decreases intimidation that students often feel towards their professors, and empowers students by providing them with positive role models and behaviors to emulate (Eagleton 2015; Lockspeiser et al. 2008). This is especially relevant in institutions where undergraduate students can apply to become TAs, and are assessed using multiple criteria, including previous academic success in the course and positive behavior when taking the course.

<table>
<thead>
<tr>
<th></th>
<th>2-yr community/technical</th>
<th>4-yr private, nonprofit</th>
<th>4-yr public, nonprofit</th>
<th>for profit</th>
<th>other</th>
</tr>
</thead>
<tbody>
<tr>
<td>Most of the time</td>
<td>7</td>
<td>20</td>
<td>20</td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>Always</td>
<td>1</td>
<td>4</td>
<td>4</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Sometimes</td>
<td>1</td>
<td>3</td>
<td>3</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>About half the time</td>
<td>1</td>
<td>2</td>
<td>2</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Never</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

*Table 6. Perceived level of student engagement with teaching assistants in anatomy and physiology classes by institution type. Categorical responses are ranked by number of responses (from high to low) at four-year public and private, nonprofit institutions (i.e., the institution category with the highest response rate to this question on the survey).*
In addition to the potential benefits to the students and the instructors, the TAs themselves receive benefits (Lachman et al. 2013). These benefits, beyond the obvious monetary compensation, may include improved, more in-depth knowledge of course material, increased capacity to communicate technical material effectively, and the opportunity to develop leadership skills. Finally, if the TAs are undergraduate students, the experience they gain can set them apart when applying to graduate schools, and the positive relationship they build with the course instructor can help them earn strong letters of recommendation for future endeavors.

Potential challenges to using TAs
While there are many benefits to having TAs in an Anatomy and Physiology course, these benefits are dependent on extensive training and oversight by the instructor, especially in the initial stages. If the selection criteria used by most instructors who participated in this survey are effective, the instructor has already chosen highly qualified, motivated individuals who are enthusiastic about teaching. However, that does not completely apply to graduate TAs who might be assigned to teach an Anatomy and Physiology lab because it is part of their graduate contract. Further study is required to determine the effect of TA assignment (i.e., competitive selection versus graduate assignment) on the engagement and success of students enrolled in Anatomy and Physiology. However, in all cases, a significant amount of time and training must be initially invested in order to have strong and competent TAs. Based on the results of the survey, formal training courses including knowledge of FERPA regulations, lab safety, and pedagogy are mainly conducted in four-year public institutions, and to a lesser degree in the four-year private institutions. In addition to formal courses, most instructors invest their own time in training TAs. This significant, and regular time investment can be potentially prohibitive, since it is likely not part of the instructor’s teaching load. The current survey did not ask how their respective departments view TA training. For example, does TA training count towards departmental service or teaching load? Once training is completed, most instructors reported having regular TA meetings throughout the semester, which is also time consuming.

Another potential difficulty is compensation. In fact, budgetary constraints were the main reason for instructors not to have TAs. The main form of compensation for institutions that utilize TAs was monetary, either through hourly wages or some form of fellowship. Justifying allocation of funds towards a TA system might not be feasible for all institutions. This seems especially relevant in community colleges, where funds are already limited, and the focus is on keeping tuition affordable for the students. However, another possibility for institutions interested in having TAs is to allow TAs to receive course credit for their work. Twenty two percent of the survey reported offering course credit to TAs as a form of compensation. Another possibility, as reported by the study participants, is offering tuition waivers.

Case Study of a Successful TA Program with Minimal Funding
One member of the present author group (Nielsen) has developed and sustained a minimally funded, but highly successful TA program that is crucial to the success of the course that reaches 500 students per semester. Attendees to the 2017 Annual Conference of the Human Anatomy and Physiology Society at the University of Utah in Salt Lake City were able to experience first-hand top-notch gross anatomy hourly workshops that were taught entirely by the undergraduate TAs of Dr. Mark Nielsen. Many faculty in attendance marveled at the preparedness of the TAs as much as the fine detail of the dissections.

In Nielsen’s course there are two large lecture sections and 12 lab sections. Thirty-six undergraduate TAs teach the labs, which constitutes an eight-to-one ratio of students to TAs. Two thirds (24) of the TAs are unpaid apprentice TAs who earned their position by being in the top five percent of the class and meeting other selection criteria. The volunteer apprentice TAs can earn course credit for the semester they teach. One third (12) of the TAs are paid, and function as experienced TAs and mentors to apprentice TAs. Graduate students requesting a TA assignment to the course must pass the same selection criteria as an undergraduate TA. The tiered structure of paid mentor and unpaid apprentice TAs not only addresses the budgetary challenge of paying large numbers of TAs, but also sustainability of training new TAs during the course as well as future course offerings. Quality teaching is maintained through an extensive, rigorous training regime that utilizes frequent meetings of mentors with apprentices. For example, if an apprentice is not prepared to teach, they will have additional meetings with their mentor. Longevity of the TA program (33 years) and the number of interested applicants each year (90-100) indicate that the program is successful. Instructor commitment to recruiting, training, and cultivating a volunteer-to-mentor TA pipeline is critical to program viability, but Nielsen credits the mentors with helping facilitate the process.

Conclusion
Considering the many benefits and challenges associated with having TAs in the Anatomy and Physiology, using TAs should be an important consideration in designing a new course or considering curriculum changes to an existing course. We hope the results of this survey provide a better perspective on how Anatomy and Physiology instructors, from a range of institution types, view the role of TAs in their classrooms and labs. Furthermore, we provide a summary of a highly respected TA system developed to address the challenges of compensation for TAs and facilitating the time-consuming training process for instructors.
Acknowledgements
We are thankful to Dr. Richard Griner (Chair, Dept. of Biological Sciences, Augusta University) and the Office of First Year and Second Year experiences for providing financial support (S. Mukhopadhyay); The University of Mississippi Office of Research and Sponsored Programs for providing access to Qualtrics survey software; Elizabethtown College for financial support (A. Goldina); HAPS for funding gift cards for survey incentives; N.M. Sanyal (Augusta University) for help with the poster presentation at the 2018 HAPS Annual Conference; and Candi Heimgartner (University of Idaho) and Melaney Farr-Birdsong (Salt Lake Community College) for help with the panel discussion at the 2017 HAPS Annual Conference. Finally, thank you to all the teaching assistants that inspired us to write this paper.

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Literature cited


Appendix A

Survey Questions for the HAPS Assessment of Teaching Assistant use in Anatomy and Physiology Courses.

Survey Flow

Description
This survey is intended to provide the members of the Human Anatomy and Physiology Society (HAPS) several key pieces of data regarding the use of teaching assistants (TAs) for the instruction of Anatomy and Physiology. We encourage HAPS members to use this data when working with their institutions to propose, develop, and cultivate programs for training TAs for quality instruction in the Anatomy and Physiology curriculum.

Cost and Payments
We estimate that it will take you approximately 15-20 minutes to complete this survey. Upon completion you may enter a raffle for one of five Amazon gift cards, one for $100 and four for $50, as our thanks for completing the survey. Winners of the raffle will be selected on November 15, 2017.

Risks and Benefits
We do not think that there are any risks to completing this survey. We anticipate publishing the results of this survey in the society’s journal, HAPS Educator, so that all members can use this information in the development and instruction of Anatomy and Physiology courses at their home institutions.

Confidentiality
No identifiable information will be recorded from the first portion of the survey. If participants wish to be entered into a raffle for one of five Amazon gift cards, one for $100 and four for $50, they will be automatically redirected to a new link to enter their email address. Participants’ email addresses will not be stored with, or linked to, responses to the first portion of the survey.

Right to Withdraw
You do not have to take part in this survey and you may stop participation at any time. If you start the survey and decide that you do not want to finish, all you have to do is to exit the survey.

IRB Approval
This study (Protocol #18x-061) has been reviewed by The University of Mississippi’s Institutional Review Board (IRB) and has been approved as Exempt under 45 CFR 46.101(b) (#2). If you have any questions, concerns, or reports regarding your rights as a participant of research, please contact the IRB at (662) 915-7482 or irb@olemiss.edu.

Statement of Consent
I have read and understand the above information. By completing the survey, I consent to participate in the study.

By checking this button, I certify that I am 18 years of age or older.

Your institution is classified as a:

- 2-yr community/technical
- 4-yr private, nonprofit
- 4-yr public, nonprofit
- for profit
- Other

On average, how many Anatomy and Physiology LECTURE sections are offered at your institution during a typical semester? Please enter a whole number rather than a range.

On average, how many students do you teach in your average Anatomy and Physiology LECTURE section? Please enter a whole number rather than a range.
On average, how many Anatomy and Physiology LAB sections are offered at your institution during a typical semester? Please enter a whole number rather than a range.

On average, how many students are enrolled in an average Anatomy and Physiology LAB section at your institution? Please enter a whole number rather than a range.

Do you use teaching assistants for Anatomy and Physiology instruction? If you select “yes”, you will be redirected to remaining questions in the survey for individuals who use TAs. If you select “No...” or “Other”, you will be redirected to a remaining question for individuals who do not use TAs.

Yes
No, but I would like to do so.
No and I do not wish to have TAs.
No, but other colleagues in my department have TAs.
Other

If you do not use teaching assistants, what is the reason? Select all that apply. After you submit your answer, you have finished the content questions and will be automatically be redirected to the end of the survey where you will find a link to enter the raffle for one of five Amazon gift cards, one for $100 and four for $50.

I do not want to use TAs for instruction of my course(s)
Budget/funding limitations
FERPA concerns (i.e. there is concern about undergrads having access to the grades of other undergrads)
University requires graduate students to be teaching assistants and undergraduates may not serve
Perceived lack of quality TAs
Time constraint/overload for faculty supervisor
Other

How many years have you been using TAs? Please enter a whole number rather than a range. Please enter a whole number.

Your teaching assistants are:
undergraduate students
graduate students
both undergraduate and graduate students
Other

In which course format(s) do you use TAs? Select all that apply.
face-to-face lecture
hybrid lecture
online lecture
face-to-face lab
hybrid lab
online lab
Other

continued on next page
How much course credit do your TAs receive per academic term?
0 credit hours
1 credit hour
2 credit hours
3 credit hours
4 credit hours
Other

How are your TAs paid?
They are not paid.
Hourly wage.
Fellowship (in the beginning/end of the semester)
Other

TA/Student Ratio -- How many students is an individual TA responsible for teaching? If you have one TA for every 5 students, enter 5. Please enter a whole number rather than a range.

What is your hiring procedure? Select all that apply.
application
high grade in the course
interview
observations of positive behavior by the student when they were taking the course
references
Other

Do you have a formal training program for your TAs?
yes, there is a formal course for training TAs
yes, I train them myself (no course for TAs)
no
Other

If you have a formal training program, which of the elements below are part of your program? Mark all that apply.
FERPA student data privacy issues
Lab Safety
Pedagogy
Mentors and apprentices (experienced and novice TA structure)
Students must complete a training course for academic credit
Other

Do you feel that your students connect academically with your TAs?
Always
Most of the time
About half the time
Sometimes
Never
How do your TAs interact with students? Select all that apply.
- station work
- help the lead lab instructor during the lab whenever students need help
- staff open lab times
- tutoring sessions
- conduct practice lab practicals
- Other

What assessment related responsibilities do your TAs have? Select all that apply.
- writing lab quizzes
- grading quizzes
- writing exams or parts of exams
- grading exams or parts of exams
- writing lab practicals
- grading lab practicals
- entering grades in a learning management system gradebook
- no grading is done by my TAs
- Other

If your TAs do grading, what types of questions? Select all that apply.
- multiple choice
- short answer
- labeling
- short essay
- skills assessment
- my TAs do not do grading of any kind
- Other

How autonomous are your TAs?
- TAs assist alongside lab instructors (or additional TAs) who are always in the room
- TAs teach without an instructor in the room
- Other

What other types of responsibilities do your TAs have? Select all that apply. Upon submission of your response, you will be automatically redirected to the end of the survey where you will find a link to enter the raffle for one of five Amazon gift cards, one for $100 and four for $50.
- set up lab
- prepare chemical reagents (ex. measuring involved)
- clean up lab
- guide students through active learning exercises in lecture
- running errands (ex. pick up supplies at stockroom or local store)
- none of these are done by my TAs
- Other
The Effects of Sex, Ethnicity, and Socioeconomic Status on Student Perceptions of Case-based Learning in Anatomy and Physiology Classes

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Abstract
Research has shown that a moderate to high level of Active Learning (AL) is helpful for all students, especially for disadvantaged students. In this study we measured student perceptions of case-based learning (CBL), a type of AL, in anatomy and physiology courses at a four-year university and a community college. We compared perceptions of CBL to student final grades and several demographic factors including sex, ethnicity, and socioeconomic status (SES). We found that students with higher final grades tended to feel that CBL increased their confidence with the course material and helped them build relationships with other students. We also found that students with lower SES were more satisfied with CBL activities and vice versa. Student populations with a higher proportion of disadvantaged students may benefit from higher levels of AL, while classes with more advantaged students may benefit from a moderate level of AL with mixed learning techniques. https://doi.org/10.21692/haps.2019.022

Key words: active learning, student perceptions, socioeconomic status, case-based learning

Introduction
Introductory Anatomy and Physiology courses have some of the highest attrition rates nationwide (Hull et al. 2016), making them an important area of educational research. Although a range exists, several studies have found attrition rates near 50%, which is higher than the science-wide attrition rate of approximately 30% (Gultice et al. 2015; Chace 2014; Hull et al. 2016). One of the paradoxes of education is that while higher education is one of the fastest ways to break away from poverty and rise in socioeconomic class, students who are in the greatest need are least likely to persist in higher education (Aragon 2000; Wells 2008). Obtaining jobs in nursing and allied health is one of the major ways that people can rise from lower to middle class and introductory Anatomy and Physiology courses serve as the gateway to these jobs (Hlinka et al. 2015).

Who are disadvantaged students in higher education?
Students most likely to give up on their education (Table 1) include those with low socioeconomic status (SES) (Number and percentage… 2013; Wapole 2003), and those with low social and cultural capital; specifically, those unlikely to use social networks to their advantage and those “without cultural indicators of symbolic wealth”. The previous two groups often include minorities (Wells 2008). Students less likely to complete their education also include males since there are lower percentages of males of all ethnicities enrolling in college and earning degrees (Percent of Recent… 2016; Wells et al. 2011). Recent medical school demographic surveys have suggested that some minorities are still under-represented (Smith et al. 2015).

<table>
<thead>
<tr>
<th>Demographics of students less likely to obtain higher education</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low SES: low parent income, parent education &amp; occupation (Number and percentage…2013)</td>
</tr>
<tr>
<td>Low social capital: parents and peers have low educational expectations (Wells 2008)</td>
</tr>
<tr>
<td>Low use of college enrollment strategies such as AP courses, SAT prep, and extracurriculars (Wolniak et al. 2016)</td>
</tr>
<tr>
<td>Fewer males than females earn college degrees (Percent of Recent… 2016), especially males from disadvantaged environments &amp; ethnic minorities particularly black (Chetty et al. 2016; Autor et al. 2015)</td>
</tr>
<tr>
<td>Ethnicity: 56.5% of black students enroll in college after high school, compared to 70% white, 70% hispanic, &amp; 87% asian (Percent of Recent… 2016)</td>
</tr>
</tbody>
</table>

Table 1. Disadvantaged students in higher education
With a growing aging population increasing the need for nursing and allied health jobs, the role of the Anatomy and Physiology instructor is vital not only to the livelihoods of our students but also to the care of our population. So the crucial question is: how do we help our students succeed in Anatomy and Physiology?

What helps students stay?
Many studies have been devoted to student retention. Vincent Tinto, one of the most influential early researchers in retention said, “students who learn are students who stay” (Tinto 1999). John M. Braxton’s retention model suggests that social integration drives a student’s commitment to their institution and thus contributes to the learning process (Braxton and Francis 2018). As an illustration of that, one study from Stanford University indicates that even a brief social-belonging intervention improved academic and health outcomes of minority students (Walton and Cohen 2011).

According to Braxton and Francis (2018) students will succeed when (1) expectations are clear, consistent and high (2) when support is available (3) when there is frequent assessment and feedback and (4) when students have high quality and frequent interactions with faculty, staff and peers. The focus of this study is to examine element four because “outsiders” in higher education are helped by increased interactions with faculty (Crosling et al. 2009) and peers through a variety of methods (Graham et al. 2013).

Rationale for using Active Learning
Active Learning (AL) is defined by Braxton et al. as “students doing things and thinking about the things they’re doing” (2008). This is certainly not the only way to accomplish the goal of having high quality and frequent interactions with faculty and peers but it is one important way. AL includes a variety of specific learning approaches that range in duration and scope (Michael 2006). The strongest argument for the efficacy of AL is a meta-analysis, published in PNAS, of 225 AL studies in which the authors conclude that AL is effective because students in STEM courses with AL were 1.5 times less likely to fail and earned 6% higher grades compared to traditional counterparts (Freeman et al. 2014). The strongest argument against the efficacy of AL is that simply having an active activity does not always improve student outcomes and may actually be unhelpful. One study showed that medical students in an AL [Problem-Based Learning] course earned lower standardized test scores and another showed doctors who were trained using AL ordered more expensive/unnecessary tests (Kirschner et al. 2006; Alferi et al. 2011; Vernon and Blake 1993). Additionally, researchers surveying AL versus traditional lecture biology courses at institutions across the US saw no difference in scores on a standardized biology quiz for 33 courses that the authors suggest may be due to having poorly-designed exercises (Andrews et al. 2011). Simply using AL is not the same thing as teaching effectively as evidence exists that instructors do not always use AL techniques in the way they were designed (Andrews et al. 2011).

Therefore, the conclusion we make is that AL is effective if done well. Michael (2006) reviewing AL research in physics, chemistry, biology, and physiology provides a thorough description of what effective AL should include based on the current understanding of how we learn; we suggest that readers new to AL refer to this article. In fact, Weiman (2014) argues in PNAS that it is no longer acceptable to use traditional lecture teaching as the comparison standard and that research needs to be done on individual AL methods. It is even likely that AL, when combined with Braxton’s other factors of student success, would lead to increased student retention (Graham et al. 2013). We emphasize the importance of thoughtful planning to prepare AL exercises according to suggestions in the literature (Table 2).

<table>
<thead>
<tr>
<th>Principles of Effective Active Learning</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clearly related to topic; reduces extraneous, attention-draining processes (Van Meter 2015)</td>
</tr>
<tr>
<td>Begins with a clear statement of purpose (Van Meter 2015)</td>
</tr>
<tr>
<td>Faculty models process and ensures students understand (Michael 2006)</td>
</tr>
<tr>
<td>Activity directs attention to key concepts (Van Meter 2015)</td>
</tr>
<tr>
<td>Progress slowly, giving multiple opportunities to problem-solve (Michael 2006)</td>
</tr>
<tr>
<td>Ends with a debriefing to reinforce key ideas (Van Meter 2015)</td>
</tr>
</tbody>
</table>

Table 2. How to make an effective Active Learning activity: a non-exhaustive list

AL is an umbrella term which includes many specialized methods such as Team-based learning (TBL) and Case-based learning (CBL). We chose CBL as it lends itself to developing clinical thinking for introductory Anatomy and Physiology courses. CBL is defined as learning that is based upon the description of a patient’s problem(s), analysis and interpretation of all the relevant data obtained from history, examination, and investigations, and planning further management of the patient (Bano et al. 2015). CBL develops critical thinking and metacognition, and can also help learners improve communication and collaboration skills (Savery 2006; Khosa and Volet 2013). While previous studies have found it hard to measure improvements in performance,
several studies have shown that students prefer CBL to other types of teaching (Bano et al. 2015; Forsgren et al. 2014; Srinivasan et al. 2007). One study found that CBL improved the patient assessment skills of undergraduate nursing students, better preparing them for clinical practice (Torreda et al. 2015).

Previous research on active learning outcomes for disadvantaged students

Haak et al. (2011) compared three types of introductory Biology courses at the University of Washington: no AL, moderate AL (daily clicker questions, weekly practice exams), and high AL (little to no traditional lecture) and found that disadvantaged students were most helped in the high AL course. All students performed better, but AL disproportionately helped disadvantaged students, closing the achievement gap by 45% in the high AL course. Perhaps the reason that AL helps disadvantaged students is that they benefit from talking through problems aloud and experiencing interdependence with peers (Hettler 2015). AL has also been shown to help students perform better on critical thinking questions (Haak et al. 2011). The conclusion, therefore is that the best kind of course for disadvantaged students is one that is entirely AL. This raises a difficult issue however, since most science courses are still taught primarily as traditional lecture and it would be problematic to require instructors to change every single meeting. Preparing and using AL can be time and labor intensive since AL should be well designed and connected to an educational philosophy that benefits students (Andrews et al. 2011). Usually studies of AL’s efficacy are done with the few best instructors at an institution (Eddy and Hogan 2014) who have been trained in educational research (Andrews et al. 2011). Since research findings at one institution may not be transferable in other contexts (Eddy and Hogan 2014), the NSF supports even repeating studies in different contexts (Widening Implementation and… 2013). Therefore, the aim of our study was to examine the effects of a low to moderate amount of AL, something doable for new instructors, on disadvantaged students.

One fourth of the higher education institutions are community colleges, and they educate one third of the students enrolled in higher education in the US (Wells 2008). Students who attend community colleges are more likely to be disadvantaged students (see Table 1) (Wells 2008). So it is fitting that research done with the goal of helping the disadvantaged in higher education be done not only at a university but also a community college. The NSF has recognized the value of community colleges and has recently provided a grant to encourage educational research in this setting (CAPER 2018).

Therefore, the goal of this study was to compare student perceptions of CBL across several demographics at a university and community college.

Methods

Four 50-minute class periods that had been traditional lectures were replaced with CBL in a second semester introductory (100-level) Anatomy and Physiology course at two institutions, a community college and a university. Two CBL cases were obtained for free from the National Center for Case Study Teaching in Science. The other two CBL cases were diagnostic games developed by the authors (Birk 2015). Since the other class periods were almost entirely traditional lecture, this is considered more than low-structure but less than moderate-structure AL (Andrews et al. 2011; Haak et al. 2011). We considered the Anatomy and Physiology lectures in both institutions to be low/moderate-structure AL. The classes at both institutions had required concurrent labs that were considered high-structure AL with multiple active learning components during each lab period.

Data were obtained during three semesters: fall 2015 through fall 2016, at two institutions: a public community college and a private four-year university. Data were collected in the form of student surveys given at the end of each semester. The surveys gathered demographic information, as well as information about student perceptions of the CBL in the form of nine Likert scale questions.

This study was approved by the Ivy Tech Community College Institutional Review Board, Protocol #15019. The research was determined to be exempt from further IRB review under Exempt Category 1: research conducted in established or commonly accepted educational settings. This study was also approved by the Southern Adventist University Institutional Review Board, IRB Tracking Number 2015-2016-007. Participation was voluntary and informed consent was obtained from all participants.

For the Anatomy and Physiology case studies, students were assigned to work in groups of five (Parmelee et al. 2009). A study done by St. George’s University used modified Subjective (chief complaint), Objective (measurements and tests done), Assessment (diagnostic), and Plan (treatment and follow-up) (SOAP) notes, to help guide small group discussions in a Medical Physiology course (Kibble et al. 2006). SOAP notes are clinical notes that medical and mental health practitioners sometimes use to document a patient’s visit and treatment on his/her medical chart. In order to make the cases in this study effective, SOAP notes were continued on next page
used to direct attention to the main concepts and to tie the case studies together in the two Anatomy and Physiology courses (Van Meter 2015; Michael 2006). Students were given examples of SOAP notes to guide them in the process (Blake 2011; Ball and Murphy 2008). Students were given pre-class reading assignments and quizzes at the beginning of class to ensure they were prepared (see Table 3).

<table>
<thead>
<tr>
<th>CBL structure</th>
<th>(10pts total per CBL, out of 1000 points in the course)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pre-class reading assignment given at the end of previous class</td>
<td></td>
</tr>
<tr>
<td>4 question Quiz given at the beginning of session covering the reading assignment (4pts)</td>
<td></td>
</tr>
<tr>
<td>SOAP notes made during class and submitted at the end (3pts)</td>
<td></td>
</tr>
<tr>
<td>Peer evaluation. An average was made of all peer evals (3pts)</td>
<td></td>
</tr>
</tbody>
</table>

Table 3. Assessments used for each AL (CBL) activity. Four 50-min. lecture periods were replaced with CBL in the semester.

The surveys included modified Likert-scale statements that connected the SOAP notes, case studies, and diagnostic games to Satisfaction, Knowledge, and Peer Interactions (see statements in Table 5).

Likert-scale statements for each of the three elements above were stated both positively and negatively. This was done to prevent students from being fed positive answers and to prevent students from filling out the same number for every answer (see statements in Tables 7 and 8). We modified the Likert scale to include a neutral as follows: 1= strongly disagree, 2= disagree, 3= neither agree nor disagree, 4= agree, 5= strongly agree.

Informed consent was obtained by verbal explanation. A student handout described the voluntary nature of the surveys and stated that participation would have no impact on a student’s grade. Faculty were required to leave the lab while students completed their surveys. A student volunteer gathered all of the survey papers and delivered them to the Department Office where the receptionists placed the surveys in a locked cabinet until after final course grades were submitted for each semester.

Statistical Methods
Analysis of the Likert data was facilitated by the use of factor analysis. This type of analysis condenses correlated variables into a single variable called a factor. This allowed us to reduce the number of variables and determine the underlying structure of the data. Of particular interest was whether the way we classified the questions was reflected in the way the students responded to them. The factor analysis was performed using Principle Component Analysis, Varimax rotation, and Keiser normalization. Factors were retained if their eigenvalues were greater than one.

Once the factor analysis was completed the factor scores were used as the dependent variables in several linear models. The dependent variables included in each model were SES score (numeric), final grade (numeric), sex (factor), and ethnicity (factor) as well as the interaction between sex and ethnicity. We did not include the type of educational institution (public community college vs. four-year university) as a separate variable in these models since: 1) preliminary data analysis (not shown) did not detect any differences between community college and four-year university students and 2) this information was largely included in the SES score since most students began their college career at the type of institution they were attending when this study was conducted (see below).

SES score was calculated by adding up the dummy variables assigned to the following demographic categories (Wells 2008): Financial Aid (0=yes, 1=no), parental degrees (0=0 parents with college degrees, 1=1 or 1 parents with college degrees), educational expectations (0= certificate or other less than associates degree, 1= expect associates or higher), peer expectations (0= none, a few, some friends expect degree, 1= most, all friends expect degree), parent expectations (0= no parent expects degree, 1= 1 or 2 parents expect degree), begin college at 2-year or 4-year institution (0=2-year, 1=4-year), begin college part or full time (0=part time, 1=full time).

We conducted statistical analysis using SPSS v. 24 (IBM Corp., Armonk, NY, USA), with alpha for hypothesis testing set at 0.05. For each linear model, we computed effect sizes as partial eta-squared ($\eta^2$). Partial eta-squared can be interpreted as percent of variance explained with values of ~0.01, ~0.06, and ≥ 0.14 corresponding loosely to small, moderate, and large effects, respectively (Cohen 1988).
The Effects of Sex, Race, and Socioeconomic Status on Student Perceptions of Case-based Learning in Anatomy and Physiology Classes

Results

General Results
Eighty-one students completed the survey. See Table 4 for student demographics and Table 5 for average Likert scores to each question. The total attrition rate at both institutions for the three semesters of second semester Anatomy and Physiology course was 7.03% (9 out of 128 students received a D, F, or W), which is lower than the nationwide science attrition rate of 30% or the approximate 50% attrition rate for Anatomy and Physiology courses (Gultice et al. 2015; Chace 2014; Hull et al. 2016).

### Table 4. Demographics information on students who completed the case-based learning (CBL) survey. N=81

<table>
<thead>
<tr>
<th>Sex</th>
<th>Male</th>
<th>Female</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age Group</td>
<td></td>
<td></td>
</tr>
<tr>
<td>18-22</td>
<td>24</td>
<td>57</td>
</tr>
<tr>
<td>23-27</td>
<td>66</td>
<td>9</td>
</tr>
<tr>
<td>28-32</td>
<td>9</td>
<td>3</td>
</tr>
<tr>
<td>33-37</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>37+</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>Ethnicity</td>
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<td></td>
</tr>
<tr>
<td>Asian</td>
<td>10</td>
<td>0</td>
</tr>
<tr>
<td>Black</td>
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<td>0</td>
</tr>
<tr>
<td>White</td>
<td>45</td>
<td>0</td>
</tr>
<tr>
<td>Hispanic</td>
<td>19</td>
<td>0</td>
</tr>
<tr>
<td>Pacific Islander</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Other</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>School</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Two-Year</td>
<td>24</td>
<td>0</td>
</tr>
<tr>
<td>Four-Year</td>
<td>57</td>
<td>0</td>
</tr>
<tr>
<td>SES Score</td>
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<td></td>
</tr>
<tr>
<td>0</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>1</td>
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<td>6</td>
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<tr>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>3</td>
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<tr>
<td>4</td>
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<td>16</td>
</tr>
<tr>
<td>6</td>
<td>7</td>
<td>11</td>
</tr>
<tr>
<td>Final Grade</td>
<td></td>
<td></td>
</tr>
<tr>
<td>A</td>
<td>22</td>
<td>0</td>
</tr>
<tr>
<td>B</td>
<td>32</td>
<td>0</td>
</tr>
<tr>
<td>C</td>
<td>25</td>
<td>0</td>
</tr>
<tr>
<td>D</td>
<td>2</td>
<td>0</td>
</tr>
</tbody>
</table>

### Table 5. Mean and SD of five-point Likert scale questions measuring student perception of case-based learning (CBL) with “strongly disagree” coded as 1 and “strongly agree” coded as 5. N = 81

<table>
<thead>
<tr>
<th>Question</th>
<th>Mean</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. The doctor diagnosis game in class helped me get to know my classmates.</td>
<td>3.74</td>
<td>0.88</td>
</tr>
<tr>
<td>2. Being familiar with SOAP notes has helped me to model the clinical reasoning process.</td>
<td>4.17</td>
<td>1.00</td>
</tr>
<tr>
<td>3. I feel that using guided case studies helped me feel more confident about class material.</td>
<td>4.01</td>
<td>0.78</td>
</tr>
<tr>
<td>4. I feel the doctor diagnosis game was a waste of my time.</td>
<td>1.98</td>
<td>1.05</td>
</tr>
<tr>
<td>5. Being familiar with SOAP notes has helped focus our group discussion</td>
<td>4.02</td>
<td>0.76</td>
</tr>
<tr>
<td>6. I feel that using guided case studies in class helped me get to know my classmates.</td>
<td>3.47</td>
<td>1.06</td>
</tr>
<tr>
<td>7. Being familiar with SOAP notes has been a useful review tool before examinations.</td>
<td>3.78</td>
<td>0.96</td>
</tr>
<tr>
<td>8. I feel the doctor diagnosis game helped me feel more confident about class material.</td>
<td>3.95</td>
<td>0.79</td>
</tr>
<tr>
<td>9. I feel that using guided case studies was a waste of my time.</td>
<td>1.84</td>
<td>0.95</td>
</tr>
</tbody>
</table>
**Factor Analysis**

Four factors were retained that grouped together the Likert questions assessing student perceptions of the SOAP notes and other AL strategies. The manner in which the analysis grouped these Likert questions is shown in Table 6. This analysis suggested that the way students responded to these questions did not completely correspond to our original question classifications. Both factor one (26.64% of the variance) and factor two (22.47% of the variance) were associated with questions related to knowledge and peer interaction. This suggested that these were closely associated in the minds of the students and that students who felt that the AL aided in their knowledge also felt that these activities increased their interaction with peers.

Factor three (14.66% of the variance) mostly confirmed our original classification as it was only associated with questions related to satisfaction. However, one of these satisfaction questions was also strongly associated with factor one and its negative loading factor suggested that students who felt that the AL strategies increased their knowledge and peer interaction generally did not feel that the doctor diagnosis game was a waste of time. Factor four (14.58% of the variance) also generally confirmed our original classification as it was largely associated with the single question on group discussion.

<table>
<thead>
<tr>
<th>Question</th>
<th>Classification</th>
<th>Factor 1</th>
<th>Factor 2</th>
<th>Factor 3</th>
<th>Factor 4</th>
</tr>
</thead>
<tbody>
<tr>
<td>Being familiar with SOAP notes has helped me to model the clinical reason...</td>
<td>Knowledge</td>
<td>0.892</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>I feel that using guided case studies in class helped me get to know my classmates.</td>
<td>Peer Interaction</td>
<td>0.661</td>
<td>0.556</td>
<td></td>
<td></td>
</tr>
<tr>
<td>I feel that using guided case studies helped me feel more confident about class material.</td>
<td>Knowledge</td>
<td>0.545</td>
<td></td>
<td></td>
<td>0.421</td>
</tr>
<tr>
<td>The doctor diagnosis game in class helped me get to know my classmates.</td>
<td>Peer Interaction</td>
<td>0.542</td>
<td>0.560</td>
<td></td>
<td></td>
</tr>
<tr>
<td>I feel the doctor diagnosis game was a waste of my time.</td>
<td>Satisfaction</td>
<td>-0.706</td>
<td></td>
<td></td>
<td>0.577</td>
</tr>
<tr>
<td>Being familiar with SOAP notes has been a useful review tool before examinations.</td>
<td>Knowledge</td>
<td></td>
<td></td>
<td>0.788</td>
<td></td>
</tr>
<tr>
<td>I feel the doctor diagnosis game helped me feel more confident about class material.</td>
<td>Knowledge</td>
<td></td>
<td></td>
<td>0.785</td>
<td></td>
</tr>
<tr>
<td>I feel that using guided case studies was a waste of my time.</td>
<td>Satisfaction</td>
<td></td>
<td></td>
<td></td>
<td>0.896</td>
</tr>
<tr>
<td>Being familiar with SOAP notes has helped focus our group discussion-</td>
<td>Group Discussion</td>
<td></td>
<td></td>
<td></td>
<td>0.927</td>
</tr>
</tbody>
</table>

**Table 6.** Principle component analysis loadings and original classifications for the Likert questions assessing student perceptions of the case-based learning (CBL) exercises. Values less than 0.4 are omitted.
The Effects of Sex, Race, and Socioeconomic Status on Student Perceptions of Case-based Learning in Anatomy and Physiology Classes

**Linear Models**
The results of this analysis are shown in Table 7. Two significant relationships were detected here. The first was that students with higher final grades tended to report that the guided case studies and SOAP notes increased confidence in their knowledge and that it helped them get to know their classmates (Factor 1, $F(1,46) = 5.06, p = 0.029$, Partial $\eta^2 = 0.10, \beta = 0.55$). The second was that higher SESs were associated with greater dissatisfaction and feelings that these educational strategies were a waste of time. The reverse is also true: lower SES students reported stronger feelings that the cases were not a waste of their time (Factor 3, $F(1,46) = 4.48, p = 0.040$, Partial $\eta^2 = 0.09, \beta = 0.20$).

![Table 7](image)

**Discussion**

**Difference by performance**
Students with higher final grades felt more confident in their knowledge of the course material and that the cases helped them get to know their classmates. Although we cannot definitively say what the cause of this correlation is, it is likely that students who earn higher grades are already the kind of students who are looking to be involved, determined, and committed to their education, reaping multiple benefits. It seems likely that students who earn high grades would be predisposed with an eagerness to get to know their classmates and those earning low grades may be less interested. One implication of this finding may be to scaffold the importance of peer interactions by presenting early in the semester that studies show that students who have high quality peer and faculty interactions are more likely to stay in higher education (Braxton et al. 2008; Crosling et al. 2009; Graham et al. 2013). It is also likely that students earning a high grade have a positive bias towards the course in that they may feel more confident in their knowledge and with the relationships they made in the class simply because they were happy with their grade.

**Satisfaction**
The correlation indicating that students with lower SES were less likely to feel that AL activities were a waste of time may indicate that intentionally using guided case studies may be better received by disadvantaged students. This is consistent with the finding that disadvantaged students benefit from an interdependence with their peers and explaining their...
answers aloud, while advantaged students are better helped by writing answers down (Hettler 2015). An increase in self-reported satisfaction with low/moderate amount of AL is a promising sign as Braxton and Francis (2018) showed that when students feel satisfied with a class they are more likely to stay in higher education. Since low SES students are at a higher risk for leaving higher education (Number and percentage... 2013; Wapole 2003), adding CBL may be a good start for instructors wanting to help disadvantaged students succeed.

The finding that high SES students were more likely to feel that AL was a waste of time seems consistent with the report that those who have high social and cultural capital, often those with high SES, feel entitled to higher education (Wells 2008), a kind of “spoiled brat” effect. The implications of this may depend on the instructor’s goals and the student population. As previously discussed, research on the efficacy of AL in general has concluded that AL is helpful for all students if done well (Freeman et al. 2014; Michael 2006; Andrews et al. 2011). AL is disproportionately helpful for disadvantaged students (Haak et al. 2011; Hettler 2015). For institutions that attract primarily disadvantaged students, such as community colleges, having more AL would be better. But for institutions that have primarily advantaged students or a broad range, perhaps using a moderate amount of AL (a few daily and weekly AL activities) would be best. We suggest a variety of techniques for such an audience as writing out answers benefits disadvantaged students, while discussing aloud benefits advantaged students (Hettler 2015).

**Future studies**

As shown by Braxton et al. (2008), when students report more positive feelings toward a course, they are more likely to be committed to their education and stay at their institution. Such studies were done at four-year residential institutions which include inherent social integration components that lead to student retention (Braxton et al. 2008), so it is a promising sign that this improvement in the satisfaction of low SES students was seen both at the university as well as the community college. Future studies should verify if student perceptions lead to student retention and increased graduation rates across higher education.

**Limitations**

There are a number of limitations to this study that should be taken into consideration. As pointed out by Hull et al. (2016), these findings rely on student perception data, which is subjective and may not reflect their ability to learn and retain knowledge of the material. Additionally, it is likely that a positive bias may exist for the students who completed the survey. Out of 128 total students enrolled in Anatomy and Physiology during the study period (including nine who received a D, F, or W), only 81 filled out the survey. The orientation toward group discussion, peer interactions, etc. of the missing students might change these results.

**Conclusion**

The two key findings were: (1) low SES students were more satisfied with the course while the opposite was also true and (2) higher grades were associated with greater confidence in student knowledge and positive peer interactions. From these findings, we suggest that instructors may want to consider the demographics of their student population when designing their courses; aiming to give student populations with a higher proportion of disadvantaged students higher levels of AL, while giving populations with more advantaged students a moderate level of AL. In the classroom we recommend incorporating a variety of AL techniques to benefit all students such as brief writing (e.g., minute essay) or other individual assignments that target students who benefit from independence as well as paired or group discussions (e.g., think-pair-share, pre-assigned teams for cases) to target students who benefit from interdependence. Outside of class we recommend a targeted approach for lower SES students. This could include offering additional peer- or TA-led AL activities such as those found on the HAPS listserv. With high attrition rates in Anatomy and Physiology courses and the growing demand for nursing and allied health professionals, we as instructors should consider designing AL activities that will help our students succeed.

**About the Authors**

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Valerie Lee, MS, taught anatomy and physiology and biology as a full-time faculty member in the biology department at Southern Adventist University in Tennessee for almost six years.

Aaron Corbit, PhD, is currently a full-time faculty member in the biology department at Southern Adventist University in Tennessee where part of his teaching load includes anatomy & physiology.

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Reference List


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The Effects of Sex, Race, and Socioeconomic Status on Student Perceptions of Case-based Learning in Anatomy and Physiology Classes


National Center for Case Study Teaching in Science. Available from: http://sciencecases.lib.buffalo.edu/cs/


Number and percentage distribution of spring 2002 high school sophomores, by highest level of education completed, and socioeconomic status and selected student characteristics while in high school: 2013. National Center for Education Statistics. Table 104.91. Available from: https://nces.ed.gov/programs/digest/d17/tables/dt17_104.91.asp


continued on next page


Diversity and Inclusion in Anatomy and Physiology Education, Degree Programs, and Professional Societies

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2School of Graduate Studies in the Health Sciences, University of Mississippi Medical Center
3School of Medicine, University of Mississippi Medical Center
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Abstract
Given the ever-increasing diversity of both the global and national community, diversity and inclusion are critical topics for consideration in healthy organizational settings. Promotion of diversity and inclusion is especially important for institutions of higher learning since diverse and inclusive environments more accurately teach learners about the real world, provide impetus for social change, and improve individuals’ skills of cross-cultural interaction. The benefits of diversity and inclusion can be experienced in the anatomical sciences, including human anatomy and physiology, when course content has relevant connections to topics of diversity and when these topics are in alignment with standard learning goals or competencies. While some organizations, including anatomy and anatomy and physiology educators, have established policies, standards, or objectives for diversity and inclusion, there are many other professional societies relevant to anatomy and/or physiology that have not adopted such guidelines. Anatomy and anatomy and physiology educators also benefit from a more diverse working environment, multicultural interactions with other individuals at their institutions or professional meetings, and diversification of anatomy instruction in the classroom with effective technology and teaching methods. The applications of diversity and inclusion foster more culturally educated and enriched global societies and anatomy and physiology communities. https://doi.org/10.21692/haps.2019.012

Key words: diversity, inclusion, human anatomy and physiology, anatomical sciences, anatomy education

Introduction
Diversity and inclusion have become popular topics in institutional climates. These topics have been important for the professional development of administrators and employees in many companies and other places of employment. According to Hays-Thomas and Bendick (2013), at least 50% of large entities boasting more than one hundred workers within the United States have diversity and inclusion initiatives and spend as much as $10 billion per year on diversity and inclusion programming. Of course, this expense approximation is six years old, if not older. Today, even more employers have established diversity and inclusion departments or offices, and these spaces have proliferated, especially in institutions of higher learning. Aguirre and Martinez (2002) described diversity as “a social force in higher education that promotes the importance of understanding difference (e.g. cultural, racial, ethnic, etc.) in building a cohesive social fabric in society” (p. 54). The authors mention the words of Baez (2000) who stressed that when learners are instructed through the lens of diversity, they come to understand the global community, identify the areas in which equity can be improved, and acquire the skills necessary to resolve the barriers to equity for the world’s citizens. Organizational communities of diverse individuals also have more leverage in contesting social hierarchies established by society (Campbell 2000) and in providing constructive feedback and critiques (Aguirre and Martinez 2002) for institutional leadership and policies. Arguably, the college or university campus is one of the best settings to foster interactions among its diverse population of students, faculty, and staff. In fact, this cosmopolitan community is perhaps one of the most effective groups in effecting change in the greater community locally, regionally, nationally, or even globally through education, research, and the exchange of ideas.

Diversity and inclusion topics do not have to be confined only to the workplace, the workforce, and the campus environment at large. They can also be integrated into course curricular standards as well as the standards of professional societies specific to certain disciplines or fields. These types of standards can even be integrated into the anatomical sciences, such as human anatomy and physiology, so that they encourage and guide faculty in incorporating diversity and inclusion activities into their classrooms. They can also be implemented in professional societies, including those that govern undergraduate education, medical education, and specifically the anatomical and physiological sciences. This paper explains some ways in which diversity and inclusion activities can be included in anatomy classrooms and...
coursework, through “hot topic” themes. It also discusses the organizations that already have diversity and inclusion standards or guidelines as well as those that could further develop such standards. And finally, it addresses the current status of female and minority representation in the anatomical sciences and the importance of increasing and improving diversity and inclusion efforts in all of these areas.

**Diversity in the Classroom**

Although racial and cultural diversity are increasing in institutions, inclusion does not necessarily always occur naturally. Humans have the tendency to interact with others with whom they share common characteristics, so institutions may have to use strategies that create more heterogeneous groups (Tienda 2013, McGlynn 2016) by challenging stereotypes (Crisp and Turner 2011). Stereotypes often serve as hindrances to diversity and inclusion efforts as preconceived notions or misconceptions that individuals have about entire groups might limit their willingness to interact with members of those groups. Thus, one course of action that can potentially dismantle or disarm such stereotypes simply involves exposing individuals to people with varying races, cultures, or ethnicities. For instance, teachers in the classroom can proactively organize their students into mixed groups that exhibit diversity (culturally, ethnically, racially, etc.). In fact, one study showed that such diverse groups improved students’ skills in interacting with other students unlike themselves simply because the courses incorporated assignments that involved heterogeneous group work and readings on topics of diversity (Engberg and Hurtado 2011). Such instances of inclusion have the potential to foster learning gains in students (Tienda 2013). Nevertheless, some more homogeneous institutions, organizations, or classrooms might not exhibit a highly diversified community. In such cases, discussions, seminars, and/or activities are extremely valuable methods for introducing diversity and fostering an inclusive mindset. Therefore, there is a need to connect curricular activities to diversity and inclusion themes.

**Types of Diversity**

There are many different kinds of diversity. However, the various forms of diversity can be organized into basic themes or categories, including demographic diversity, experiential diversity, and cognitive diversity (de Anca and Aragón 2018). Table 1 provides a list of specific types of diversity organized under these more general categories, but this list is by no means an exhaustive list of all of the many forms of diversity. The discussion of these various forms of diversity is important as they appeal to individuals’ identities of origin, growth, and aspiration (de Anca and Aragón 2018). Highlighting these identities celebrates diversity and creates an environment of cultural awareness, sensitivity, and appreciation that promotes inclusion. The themes encompassing these types of diversity can be applied even to a field as specific as anatomy, through their application to “hot topics.”

<table>
<thead>
<tr>
<th>Categories, or Themes</th>
<th>Identities</th>
<th>Types</th>
</tr>
</thead>
<tbody>
<tr>
<td>Demographic diversity</td>
<td>Origin</td>
<td>Age, Biological sex, Ethnicity, Gender, Race, Sexual orientation</td>
</tr>
<tr>
<td>Experiential diversity</td>
<td>Growth</td>
<td>Abilities, Affinities, Cultural, Disabilities, Hobbies, Nationality, Political views, Religious, spiritual, or secular views, Socioeconomic status</td>
</tr>
<tr>
<td>Cognitive diversity</td>
<td>Aspiration</td>
<td>Education, Learning preferences, Occupation, Personality, Spatial abilities</td>
</tr>
</tbody>
</table>

**Table 1. Types of Diversity.** This table display various types of diversity according to three general themes or categories of diversity and to three basic identities to which individuals ascribe. These themes and identities were assembled from an article by de Anca and Aragón (2018). The types have been assigned to them by the authors of this paper, but they do not represent an exhaustive list. A number of these types have been explored in the field of diversity and inclusion management (Theodorakopoulos and Budhwar 2015).
Diversity in Anatomy Education

This section explores three general themes of diversity and provides general ideas for specific examples of how they can be incorporated into classroom activities that pertain to relevant topics in the fields of anatomy and human anatomy and physiology.

1. Demographic Diversity
The theme of demographic diversity perhaps incorporates more types of diversity compared to the other themes mentioned in this paper. While excess detail is not allocated to the discussion of examples of each of the types outlined in Table 1, one example of how content pertaining to gender diversity can be incorporated into the classroom is described. The topic of gender diversity is arguably the hottest current topic in anatomy, especially given the controversy that discussions of gender have instigated in society. In fact, at the Experimental Biology Annual Meeting in San Diego, California, on April 21, 2018, the American Association of Anatomists (AAA), now known as the American Association for Anatomy (AAA), featured a symposium titled “The Anatomy of Gender” (Carter et al. 2018). In this symposium, two anatomists Drs. Yasmine Carter and Derek Harmon discussed the importance of including in medical curricula terminology pertaining to transgender and gender-nonconforming individuals, such as appropriate pronouns and terminology to use in patient interactions. In the same symposium a urologist, Dr. Maurice Garcia, described in detail the steps taken to conduct feminizing and masculinizing surgeries on patients desiring to undergo transition. Dr. Harmon also emphasized the importance of incorporating transgender anatomy into anatomy curricula, and Dr. Carter asserted the relevance of such content to the Liaison Committee for Medical Education (LCME) standards.

Instructors in anatomy and physiology or other anatomy courses can incorporate topics similar to those discussed in this symposium. Workshops pertaining to this content have also been conducted at two medical centers in the country, where Drs. Harmon and Garcia performed pelvic procedures portraying various stages of feminizing and masculinizing surgeries and led urology and obstetrics and gynecology residents in performing the surgical procedures themselves. Although the cost would be high, a similar workshop featuring procedures could also be done at undergraduate institutions for anatomy and physiology students. For lessons involving little or no expense, instructors could simply use visual presentations with diagrams and photos, and they could lead their students in discussions regarding the current literature on transgenderism. Such activities might also be beneficial for professional students in the health sciences since a recent review article has noted substantial knowledge gaps across medical subspecialties (Wanta and Unger 2017). Educators can also refer students to literature asserting the scientific difference between gender and biological sex (Zhou et al. 1995, Torgrimson and Minson 2005, Bao and Swaab 2011, Smith et al. 2015, Feusner et al. 2017) to help clear up any misconceptions that students might have concerning this difference which has been so controversial in society. Furthermore, educators can discuss various physiological differences that exist between males and females; knowledge that has only emerged fairly recently, beginning in the mid-1990s when women began to be included in clinical trials (Torgrimson and Minson 2005).

In anatomy and physiology courses, anatomy concerning gender can be an essential topic of diversity for a number of reasons. First of all, such discussions are important for students to have, especially since they likely will meet people who identify as transgender individuals or who do not conform to a specific gender. Secondly, conversations surrounding gender increase individuals’ sensitivity to characteristics that are different from their own. In addition, gender equality and equity are important considerations in creating a more diversified representation of female, trans, and other gender non-conforming individuals between student and workforce populations at institutions of higher learning. This fact is especially true since there is a discrepancy in the higher number of female students graduating from programs in the sciences and the lower number of female faculty obtaining jobs (Snyder et al. 2019). There are probably even smaller numbers of transgender, gender non-binary, or gender non-conforming individuals entering these programs as students or faculty, but the numbers are not currently reported in statistics since federal law still requires gender to be reported as the binary designations of “male” or “female,” and since there is ambiguity as to whether the data and terminology represent gender, biological sex, or both (IPEDS 2017). The fields of anatomy and neuroscience can play instrumental roles in changing such an impractical law. Finally, since many of the students who take courses in either anatomy or physiology or combined anatomy and physiology courses ultimately intend to enroll in health professional programs, they must be aware of the varying terminology and anatomy among transgender individuals in various stages of transition as they may have interactions with such patients in the future.

Another example of content pertaining to demographic diversity involves racial diversity. For instance, given the racial demographics of Mississippi and its institutions like the University of Mississippi Medical Center (UMMC), activities of corresponding racial diversity can be included in undergraduate anatomy and physiology courses. For example, students learning about red blood cells could study the prevalence of sickle cell anemia in people of African descent and the evolutionary advantages the genetic trait offers to malaria resistance. In addition, further reading could feature the prevalence of thalassemia in Caucasians. Examples of evidence-based research might revolve around studies that analyze the predispositions of certain racial and socioeconomic groups to display health care disparities.
For instance, in Mississippi, the Jackson Heart Study (JHS), the largest investigation of causes of cardiovascular disease in African Americans, has shown a higher prevalence of hypertension, coronary heart disease, stroke, heart failure, and peripheral arterial disease in African American males than their Caucasian counterparts (HHS 2019). Furthermore, anatomy and physiology instructors could organize students into small groups whereby students might discuss the factors contributing to higher risks of diseases in particular racial or ethnic groups, such as heredity, access to healthy foods, or other factors. Therefore, if the demographics of the students in their classes are different, instructors could incorporate diversity content relevant to the ethnic, racial, or other cultural groups represented by their particular students, or they can discuss content pertaining to multicultural groups that are different from the ones represented in their classrooms. In this respect, instructors are able to integrate diversity in classrooms, even if their student cohorts lack racial or ethnic diversity.

2. Experiential Diversity

In addition to incorporating content pertaining to topics of demographic diversity, instructors can incorporate content pertaining to topics of experiential diversity into their classrooms. One such topic regarding the ethical considerations of body donation programs and the use of anatomical donors and other cadaveric materials has implications with religious, spiritual, and secular diversity and has high relevance in anatomy education. Instructors in human anatomy or human anatomy and physiology courses can address the implications when they discuss topics such as respect for human tissue or for the human as a whole (Murray 1986, Lella and Pawluch 1988, Upshur et al. 2007, Satyapal 2012). Instructors can also facilitate conversations about the treatment of the human body from the vantage point of different religious beliefs (Daar 1994, Daar 1997, Raza and Hedayat 2004, Park et al. 2011). Another emerging hot topic that can entail religious and spiritual undertones is the anxiety associated with the fear of death and dying (Howells et al. 1986, Black et al. 1989, Williams et al. 2005, Thiemann et al. 2015). Sara Klender, a colleague and current graduate student at UMMC, has begun to explore the fear of death among dental students and its impact on their performance in gross anatomy (Klender et al. 2019). She intends to conduct her dissertation research on this topic while exploring potential ways of alleviating this anxiety among both dental and medical students; many of whom confront real human donors for the first time in their gross anatomy courses. Such research can also instill within all students who work with cadavers, not just medical students, a more authentic understanding of mortality and respect for the dignity of the human body.

The two main topics that are often considered taboo for intellectual discourse are religion and politics. However, religious and political values are indeed those ideas which govern important aspects of human activity, such as the daily functioning of a college or university, legislation concerning gender equity and other forms of social justice, and the use of human specimens in the education of students in the health sciences. By having such difficult conversations, people not only learn how to formulate arguments which enable them to articulate their own diverse perspectives in an educational forum, but they also learn how to clarify misconceptions that they might have possessed regarding a particular religious belief or political ideology. Furthermore, people can develop a deeper respect for the religious or political beliefs of their colleagues or classmates by learning about them. For instance, Amberly Reynolds, a colleague and current graduate student at Indiana University, offered a workshop titled “A Native American Perspective on Medicine, Donors and Dying” at the most recent Human Anatomy and Physiology Society (HAPS) Annual Meeting. In her workshop, she presented the customs of the Navajo tribe, including the religious practices, tribal medicine rituals, and treatment of their dead. Should Navajo students work with the deceased, as in a Gross Anatomy Laboratory, they must undergo a cleansing ritual before interacting with family and crossing the home threshold. Understanding the spiritual beliefs that forbid students from handling the bodies of those who are no longer living can better prepare instructors and other students for respecting these students’ potential wishes to abstain from handling deceased human tissue.

3. Cognitive Diversity

When discussing topics of diversity in the classroom, instructors can also consider topics pertaining to cognitive diversity in addition to demographic and experiential diversity. One form of cognitive diversity that instructors might want to incorporate into their instruction is occupational diversity, given the various professions relevant to the field of anatomy. Instructors might want to survey their students to obtain a better idea of which fields many of them want to pursue. Given the fact that many undergraduate students in human anatomy and physiology or human anatomy courses tend to have an affinity for the health sciences, instructors can incorporate content pertaining to medicine, nursing, nurse anesthesia, dentistry, pharmacy, occupational therapy, physical therapy, physician assistant, optometry, and/or mortuary science programs. Additional programs that might also have some varying degrees of relevancy to anatomy include anthropology and veterinary medicine. Since each discipline places different emphases on certain regions and systems of the body and on certain clinical skills that require more application of anatomy, instructors can design their courses to make corresponding curricular accommodations for their students. A diverse and inclusive approach to all or several of these disciplines in undergraduate classrooms might also help students make informed decisions about the careers they both do and do not want to pursue upon graduation.
Another form of cognitive diversity that becomes especially important when studying anatomy both in the cadaveric lab and via computer-based virtual modalities is that revolving around individuals’ spatial abilities. In fact, several studies have demonstrated that students with high spatial abilities perform better in completing complex tasks (Yang et al. 2003, Rusch 2008, Höffler, 2010, Riastuti et al., 2017) as well as in learning anatomy (Garg et al. 2001, Luursem 2006 and 2008, Luursem and Verwey 2011, Nguyen et al. 2014, Berney et al. 2015, Cui et al. 2017) than their lower spatial ability counterparts. Anatomy instructors, therefore, should consider using instructional methods that help students with lower spatial abilities learn anatomy just as well as, if not better than, their high spatial ability counterparts. For instance, with the increase in virtual anatomy over the past few decades, there is a need to explore situations in which the use of cadavers or cadaveric material is more beneficial than virtual models or images and vice versa. Moreover, there is a need to examine situations in which two-dimensional (2D) images are more appropriate than three-dimensional (3D) images and vice versa and in which static images are more appropriate than movable images and vice versa. Furthermore, of the 3D images, instructors must consider when monoscopic and stereoscopic images are better, if at all. Monoscopic images refer to those that can be rotated in three dimensions on a flat screen while stereoscopic images refer to those that appear to “pop out” of the screen similar to a 3D movie.

A factor of spatial abilities known as spatial relation ability (SR), which refers to an individual’s capacity to rotate 2D and 3D images in their mind rapidly and accurately, has a positive correlation with improved task performances (Berney et al. 2015). Individuals utilize SR when they must form mental representations of 3D objects (Berney et al. 2015), especially when those objects possess an internal anatomy of structures with identifiable 3D relationships to each other (Meyer 2019). The mental rotation test (MRT) has been classically used to measure individuals’ SR (Shepard and Metzler 1971, Vandenberg and Kuse 1978, Peters et al. 1995), and the MRT items can be obtained from Peters et al. with permission and then used to assess students’ or other individuals’ SR. According to studies, low SR students learning with movable monoscopic (Berney et al. 2015) or stereoscopic visualizations performed just as well as high SR students (Luursem et al. 2006, Cui et al. 2017). These studies suggest that the movable visualizations accommodate low SR students (Mayer and Sims 1994, Hays 1996, Mayer 2002) but provide fewer gains for high SR students. Conversely, high SR students who learned with immovable 2D visualizations performed better than low SR students (Berney et al. 2015, Cui et al. 2017). In another study comparing two groups of first-year medical students learning from either monoscopic or stereoscopic middle and inner ear models, students with lower spatial ability had practically higher scores than their higher spatial ability colleagues, despite the lack of a significant difference between the two (Meyer 2019). Instructors can gather data on their own students’ spatial abilities and examine their performance after instructional interventions to determine which methods work optimally for their classes.

**Diversity and Inclusion Standards**

A number of national and international professional societies and organizations that include members who are anatomy or anatomy and physiology instructors have adopted diversity and inclusion standards by which to govern their interactions with a diverse membership and to promote inclusive behaviors among their members. Appendix 1 provides a list of a few of these professional societies or organizations and their contributions to diversity and inclusion. However, this appendix table is, in no way, an exhaustive list of all of the societies and organizations in the health sciences that may contain members who are anatomy or anatomy and physiology educators. Details about the organizations that are not discussed in this section can be found by consulting Appendix 1 to learn more about their contributions to diversity and inclusion. Of the 49 professional societies or organizations included in the table, approximately 10% provided a set of standards or equivalent, approximately 27% provided a statement or equivalent, approximately 27% mentioned a committee or equivalent, and approximately 18% provided an official webpage with resources dedicated to diversity and inclusion.

**Quicklink to APPENDIX 1.**

**Contributions of Anatomical and/or Physiological Societies**

Of the professional societies specific to anatomy and/or physiology, the American Association for Anatomy (AAA) and the American Physiological Society (APS) appear to make the greatest contributions to diversity and inclusion. Unlike the AAA, the APS includes a set of diversity and inclusion policies as well as a webpage dedicated to diversity and inclusion resources. The AAA does have a fairly new Diversity and Inclusion Committee unlike the APS; however, the APS has a few committees dedicated to minority groups in the field of physiology. In addition, the Physiological Society (PS) has a set of diversity and inclusion objectives as well as a webpage featuring diversity and inclusion resources. In turn, while the Human Anatomy and Physiology Society (HAPS) does not have policies, standards, or objectives that explicitly address cultural competency or diversity and inclusion like the APS, it does have a set of learning goals (HAPS 2018) for anatomy and physiology students. As a matter of fact, the organization’s broader process goals (Figure 1) provide an invitation for instructors to incorporate in their courses readings on diversity topics relevant to anatomy and physiology, anatomy and physiology research featuring diverse sample populations, and heterogeneous small-group collaborations. These course activities could exhibit any amount of variability depending on the location of the institutions where anatomy and
physiology courses are offered. Some examples of activities or opportunities for incorporating diversity and inclusion topics into the classroom were discussed in a previous section.

Contributions of Health Professional Societies

Overall, medical centers or health science centers with medical schools seem to have taken the lead in promoting diversity and inclusion in the medical profession as well as in other healthcare fields. Many medical schools in the United Kingdom, the United States, and Canada have included diversity and inclusion competencies or standards in response to increasing diversity in populations (Dogra et al. 2009). Schools that are members of the Association of American Medical Colleges (AAMC) and the American Medical Association (AMA) have curricula that are governed by the Liaison Committee on Medical Education (LCME), which includes cultural competency in its standards (Figure 2) (LCME 2016). While this standard conveys the LCME’s commitment to ensuring that medical students understand the importance of cultural diversity, there is certainly a lack of uniformity in the implementation of the standard among medical centers across the country. The AAMC and AMA are both featured in Appendix 1. In fact, through its website, the AAMC makes one of the most impressive contributions of resources devoted to diversity and inclusion (AAMC 2019). These resources can be freely accessed at any time to aid individuals in implementing or organizing diversity and inclusion programs of their own. Furthermore, the AAMC boasts diversity and inclusion as one of its missions with a Chief Diversity and Inclusion Officer and a team of other staff members under his leadership.

Broader Process Goals

“The skills included in these goals should be developed while students are taking Anatomy and Physiology, but will also be reinforced in other curricular coursework. It is recommended that assignments and projects be used that develop these skills within the context of the fundamental content and process goals (1-8, above). The HAPS Curriculum and Instruction Committee has not linked these goals to specific learning outcomes in anatomy and physiology. Rather, the goals can be adapted to fit where the instructor feels they are most appropriate within the course content. There is no expectation of equal emphasis throughout all anatomy and physiology topics.

9. Demonstrate information literacy skills to access, evaluate, and use resources to stay current in the fields of anatomy and physiology.

10. Approach and examine issues related to anatomy and physiology from an evidence-based perspective.

11. Communicate clearly and in a way that reflects knowledge and understanding of the human body and demonstrates the ability to adapt information to different audiences and applications.” (HAPS 2018)

Figure 1. Broader Process Goals of the Human Anatomy and Physiology Society (HAPS). These general goals allow the instructor room to modify his or her course learning objectives to promote not only depth of knowledge and application but also breadth of knowledge and application, perhaps for multidisciplinary connections such as diversity and inclusion.

7.6 Cultural Competence and Health Care Disparities

“The faculty of a medical school ensure that the medical curriculum provides opportunities for medical students to learn to recognize and appropriately address gender and cultural biases in themselves, in others, and in the health care delivery process. The medical curriculum includes instruction regarding the following:

- The manner in which people of diverse cultures and belief systems perceive health and illness and respond to various symptoms, diseases, and treatments
- The basic principles of culturally competent health care
- The recognition and development of solutions for health care disparities
- The importance of meeting the health care needs of medically underserved populations
- The development of core professional attributes (e.g., altruism, accountability) needed to provide effective care in a multidimensional and diverse society” (LCME 2016).

Figure 2. Liaison Committee on Medical Education (LCME) Standards for Cultural Competence. These standards are required to be included in the curricula of all medical schools that are members of the Association of American Medical Colleges (AAMC).

continued on next page
Several of the professional societies representing other healthcare disciplines in Appendix 1 make varied contributions to diversity and inclusion on their websites. The field of nursing makes notable contributions. For instance, the American Academy of Nursing (AAN), American Association of Nurse Anesthetists (AANA), and the American Association of Nurse Practitioners (AANP) all have Diversity and Inclusion committees. Although the American Association of Colleges of Nursing (AACN) does not have a committee specifically dedicated to Diversity and Inclusion, it does have a Diversity, Equity, and Inclusion Group (DEIG). In addition, diversity and inclusion resources can be found on the websites of the AACN, AANP, and ANA as well as the Physician Assistant Education Association (PAEA). Of these other health professional societies, a set of policies, objectives, or goals can be found on the websites of the American Occupational Therapy Association (AOTA), PAEA, and American College of Radiology (ACR).

Contributions of Other Professional Societies
Among the other professional societies included in Appendix 1, noteworthy contributions to diversity and inclusion are made by the American Veterinary Medical Association (AVMA) and the Palaeontological Society (PS), both of which include statements of diversity and inclusion on their website. Moreover, the AVMA posts diversity and inclusion resources, and the PS has a Diversity and Inclusion Committee. While the American Association for the Advancement of Science (AAAS) and the Federation of American Societies for Experimental Biology (FASEB) do not include content relevant to Appendix 1 on their websites, they each make notable contributions to diversity. Both the AAAS and the Association of American Universities (AAU) produced a handbook on diversity and associated legal matters (Ham 2010), and the FASEB offers several federally funded opportunities through its Diversity Resources Program (FASEB 2017). There are certainly more organizations or professional societies relevant to anatomy and/or anatomy and physiology that are not discussed in this paper.

Diversity in the United States
Since this paper mainly focuses on diversity and inclusion in anatomy education and professional development in the United States, considerations of the state of diversity in the United States as a whole are important. In this section, this article takes a closer look at the most recently reported statistics concerning racial and ethnic diversity as well as gender diversity in the United States. Many of these statistics specifically highlight the number of minorities and the number of women enrolling and graduating from anatomical and physiological programs with bachelor’s, master’s, and doctoral degrees. Unfortunately, there are no databases that report transgender, gender non-binary, and gender non-conforming individuals. For this reason, this section will describe these “male” and “female” designations as biological sex rather than gender since the actual gender of the individuals is not clear. This section also explores the racial, ethnic, and gender diversity of students and faculty in baccalaureate and post-baccalaureate institutions where courses in human anatomy and/or human anatomy and physiology might be offered.

The United States as a Whole
From 2015 to 2016, all racial and ethnic minority groups proliferated faster than the Caucasian majority, thus demonstrating that racial and ethnic diversity in the United States is steadily increasing (Chappell 2017). The Census projects that more infants of color will be born in 2020 than white infants (Yoshinaga 2016). In fact, by 2045, Caucasians will be replaced as a minority comprising 49.7% of the population while the collective minority groups will comprise the majority (50.3%) of the population, including Hispanics (24.6%), Blacks (13.1%), Asians (7.9%), and multiracial individuals (3.8%) (Frey 2018). Given these estimations, efforts in improving and increasing diverse and inclusive interactions are paramount. A meta-analysis of college diversity experiences and civic engagement has shown that students who are exposed to diversity-related activities are more likely to develop civic-minded attitudes and to engage in civic behaviors (Bowman 2011). In addition, a study including survey data from over 15,000 students from 102 different institutions showed that a healthy institutional campus climate is dependent on racial and socioeconomic diversity (Park et al. 2013). Similarly, the more racial and socioeconomic diversity are promoted on any institution’s campus, the greater the potential that the institutional climate will be improved even further. In fact, instructors who are especially passionate about diversity and inclusion might consider expanding diversity and inclusion activities beyond the anatomy and physiology or human anatomy classroom and into other areas of their institutional environments. Such efforts are extremely worthy given the underrepresentation of minority groups in baccalaureate and post-baccalaureate institutions.

Diversity in Baccalaureate and Post-Baccalaureate Institutions
This subsection presents information regarding the number of students enrolled in anatomy- and/or physiology-relevant programs in 2016 according to the most recent data from the Digest of Education Statistics 2017 (Snyder et al. 2019). It also presents the number of degrees conferred to bachelor’s, master’s, and doctoral students in these relevant disciplines in totality and then by race/ethnicity and sex in 2016. Furthermore, it presents the number of faculty employed by these institutions in totality and then by sex and race/ethnicity in 2016.

Upon general overview of the change in graduate enrollment in research-based programs in degree-granting post-secondary institutions, there has been an increase in the number of enrollees in the biological sciences over a ten-year period of time. According to the Digest of Education Statistics 2017, that number has increased from 15,898 students in 2007 to 16,380 students in 2016 (Snyder et al. 2019). In
addition, when comparing the number of enrolled students during these two years, there have been enrollment increases in physiology (2,738 vs. 4,024) and in neurobiology and neuroscience (1,584 vs. 5,226) (Snyder et al. 2019). However, there has been a noticeable decrease in the number of enrolled students in anatomy (867 vs. 448) when comparing the number of enrolled students for 2007 and 2016 (Snyder et al. 2019). This decrease in the number of students entering anatomy programs throughout the country might play a role in the paucity of new classically trained anatomists at post-secondary institutions in the United States. The need for anatomy educators is not only experienced in the United States and Canada but also in the European Union (Wilson et al. 2019). Between 2017 and 2018 at medical schools in the United States alone, anatomy job postings increased more than two-fold, yet nearly a quarter were left unfilled (Wilson et al. 2019). The demand for more anatomy educators warrants the establishment of more programs for training anatomy scholars and educators not only for the sake of supplying the demand but also for the sake of increasing the diversity of individuals within the field. If the number of individuals dwindles so too will the diversity.

The next set of tables were created from specific NCES data pertaining to the total number of anatomy- and physiology-related U.S. post-secondary degrees obtained by students, the number of those degrees according to race/ethnicity and sex, and the number of U.S. faculty according to race/ethnicity and sex (Snyder et al. 2019). Table 2 presents the total number of bachelor’s, master’s, and doctoral degrees in anatomy- and physiology-related programs according to sex conferred by post-secondary institutions at the culmination of the 2015-2016 academic year. Table 3 compares the number of bachelor’s, master’s, and doctoral degrees in the biological and biomedical sciences conferred by post-secondary institutions according to race/ethnicity and sex between the 2014-2015 and 2015-2016 academic years. Understandably, there is a discrepancy in the previously mentioned number of enrollees versus the number of graduates given the fact that not all who matriculate into degree programs graduate from them.

<table>
<thead>
<tr>
<th>Degree</th>
<th>Bachelor’s</th>
<th>Master’s</th>
<th>Doctoral</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex</td>
<td>Male</td>
<td>Female</td>
<td>Total</td>
</tr>
<tr>
<td>Field &amp; Program</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Anatomy</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Anatomy</td>
<td>157</td>
<td>257</td>
<td>414</td>
</tr>
<tr>
<td>Cell Biology &amp; Anatomy</td>
<td>2</td>
<td>3</td>
<td>5</td>
</tr>
<tr>
<td>Cell Biology &amp; histology</td>
<td>193</td>
<td>244</td>
<td>437</td>
</tr>
<tr>
<td>Cell/Cellular Biology &amp; Anatomical Sciences</td>
<td>61</td>
<td>68</td>
<td>129</td>
</tr>
<tr>
<td>Developmental Biology &amp; Embryology</td>
<td>27</td>
<td>37</td>
<td>64</td>
</tr>
<tr>
<td>Neurobiology &amp; Anatomy</td>
<td>347</td>
<td>450</td>
<td>797</td>
</tr>
<tr>
<td>Total</td>
<td>787</td>
<td>1,059</td>
<td>1,846</td>
</tr>
<tr>
<td>Physiology</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cell Physiology</td>
<td>0</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Exercise Physiology</td>
<td>1,507</td>
<td>1,810</td>
<td>3,317</td>
</tr>
<tr>
<td>Physiology, General</td>
<td>702</td>
<td>912</td>
<td>1,614</td>
</tr>
<tr>
<td>Total</td>
<td>2,209</td>
<td>2,723</td>
<td>4,932</td>
</tr>
<tr>
<td>Biological Sciences, General</td>
<td>27,889</td>
<td>45,152</td>
<td>73,041</td>
</tr>
</tbody>
</table>

Table 2. Anatomy and Physiology Degrees Conferred by Post-Secondary Institutions in the United States According to Biological Sex in the 2015-2016 Academic Year. This table represents the number of bachelor’s, master’s, and doctoral degrees conferred to both males and females and in total at the culmination of the 2015-2016 academic year (Snyder et al. 2019). The degrees are only those associated with programs in the anatomical, physiological, and general biological sciences. These data were collected from both private and public institutions.
Table 3. Biological and Biomedical Science Degrees Conferred by Post-Secondary Institutions in the United States According to Biological Sex and Race/Ethnicity in the 2014-2015 and 2015-2016 Academic Years. This table represents the number of bachelor’s, master’s, and doctoral degrees conferred to both males and females at the culmination of the 2014-2015 and 2015-2016 academic years, respectively (Snyder et al. 2019). The NCES only reported data regarding the total number of bachelor’s degrees for males and females separately. The total number of males and females receiving master’s and doctoral degrees were reported together, but not separately. Specific data regarding the race/ethnicity of enrollees in anatomy and physiology programs were also not reported by the NCES, but since anatomy and physiology are included within the biomedical sciences, the numbers of students according to sex and race/ethnicity are embedded in the data within this table. The minority total represents the sum of the number of Black, Hispanic, Asian, Pacific Islander, and American Indian/Alaskan Native students as well as the number of students who identify with two or more races. These data were collected from both private and public institutions.

Yr. = Year; Deg. = Degree; Bach. = Bachelor’s; Mast. = Master’s; Doct. = Doctoral; M = male; F = female; T = Total; W = White; B = Black; H = Hispanic; A = Asian; PI = Pacific Islander; AI/AN = American Indian/Alaskan Native; 2+ races = two or more races; Min. = Minority; N-RA = non-resident alien.
Overall, there was approximately a one-percent increase in diversity in terms of the number of minorities receiving degrees between the 2014-2015 and 2015-2016 school years. The only minority groups that experienced a decrease in the number of degrees conferred were the Pacific Islander male and female students as well as the American Indian/Alaskan Native male students. In addition, the number of White male students receiving degrees between these two academic years also decreased. Although the number of Black male students receiving degrees has increased, the rate of increase relative to the population of Black students is not very high. Further research could be aimed at exploring the reason for these decreases and low rates of increase for these respective groups.

In addition to the diversity of the students in post-secondary institutions, the diversity of the faculty is also analyzed. Table 4 presents the total number of faculty employed by post-secondary institutions in the United States in 2016 according to both race/ethnicity and sex. There has been an approximate 0.4-million decrease in the number of faculty employed by U.S. post-secondary institutions; approximately 1.5 million in 2016 and approximately 1.1 million in 2017 (Snyder et al. 2019). This decrease is likely due to the legislative cuts to government spending on education. Nevertheless, when comparing the numbers of male and female faculty between 2005 and 2016, specifically, there has been a 9.9-percent increase in male faculty (714,453 vs. 785,157) and a 32.6-percent increase in female faculty (575,973 vs. 763,575) (Snyder et al. 2019). While such percentages bode well for diversity on the basis of sex, a closer look shows that the number of male faculty still outnumber the number of female faculty by over 20,000. Moreover, in 2016, male faculty on average were paid 21 percent more than female faculty (Snyder et al. 2019). The percentage of faculty represented by females in minority groups was two points higher than that represented by males in minority groups in 2016, and the total percentage of either males or females in minority groups was still below 25%. Although these statistics describe the overall landscape of diversity among all faculty within U.S. post-secondary institutions rather than only the diversity within anatomy- and physiology-relevant disciplines, these data still strongly suggest the need for further research in the diversity of the workforce in anatomical and/or physiological fields. Plus, if a lack of diversity exists on the basis of race/ethnicity and/or sex, measures for promoting more diverse and inclusive environments within individual departments should be explored.

<table>
<thead>
<tr>
<th>R/E →</th>
<th>B</th>
<th>H</th>
<th>A</th>
<th>PI</th>
<th>AI/AN</th>
<th>2+ races</th>
<th>Min. Total</th>
<th>N-RA</th>
<th>U/U</th>
<th>W</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>M</td>
<td>42,018</td>
<td>36,905</td>
<td>62,288</td>
<td>1,834</td>
<td>3,408</td>
<td>6,420</td>
<td>152,873</td>
<td>34,772</td>
<td>36,938</td>
<td>560,574</td>
<td>785,157</td>
</tr>
<tr>
<td>F</td>
<td>63,598</td>
<td>37,762</td>
<td>50,812</td>
<td>1,903</td>
<td>3,807</td>
<td>7,710</td>
<td>165,592</td>
<td>20,815</td>
<td>38,510</td>
<td>538,658</td>
<td>763,575</td>
</tr>
<tr>
<td>T</td>
<td>105,616</td>
<td>74,667</td>
<td>113,100</td>
<td>3,737</td>
<td>7,215</td>
<td>14,130</td>
<td>318,465</td>
<td>55,587</td>
<td>75,448</td>
<td>1,099,232</td>
<td>1,548,732</td>
</tr>
</tbody>
</table>

Table 4. Faculty Employed by Post-Secondary Institutions in the United States in the 2015-2016 Academic Year According to Race/Ethnicity and Sex. This table represents the number of faculty in all degree programs according to race/ethnicity and biological sex (Snyder et al. 2019). The NCES did not report data on faculty for each specific degree type or program. The minority total represents the sum of the number of Black, Hispanic, Asian, Pacific Islander, and American Indian/Alaskan Native faculty members as well as the number of faculty members who identify with two or more races. These data were collected from both private and public institutions.

R/E = Race/Ethnicity; M = Male; F = Female; T = Total; B = Black; H = Hispanic; A = Asian; PI = Pacific Islander; AI/AN = American Indian/Alaskan Native; 2+ races = two or more races; Min. = Minority; N-RA = non-resident alien; U/U = Unknown/Unreported; W = White
Fostering Diversity and Inclusion Within and Beyond the Classroom

Whether the settings are classrooms, departmental workspaces, conference seminars or meetings, or institutional offices or buildings, similar practices for promoting diversity and inclusion can be implemented in any environment. As long as personnel embrace ways of maintaining and further improving diversity and inclusion, diverse and inclusive atmospheres can flourish. The literature has revealed that diversity and inclusion are not the same and that promoting diversity within any group without also fostering inclusion is a moot point (Stevens et al. 2008, Stewart et al. 2008, Tienda 2013, Ferdman and Deane 2014, Sabharwal 2014, Ferdman 2017). Diversity and inclusion involve celebrating individuals’ difference while simultaneously allowing individuals to feel a part of a group sharing in a common mission. The key then to authentic diversity and inclusion is finding the right balance in providing group members the often contradictory realities of a sense of belonging and the allowance of self-expression of unique identities (Ferdman 2010, Shore et al. 2011, Ferdman 2017). Any group can obtain this balance.

Within the Classroom

In addition to expanding and elaborating on the classroom diversity activity examples provided earlier in this paper, instructors of anatomy or anatomy and physiology can establish inclusive classrooms in several ways. For instance, to foster inclusion, instructors can build rapport with their students through open conversation at the beginning and ending of class sessions, encourage student interactions by incorporating icebreakers into lessons, and facilitate small-group activities involving diverse student members (McGlynn 2016). Instructors can also utilize inclusive assessment methods to ensure a safe, equitable environment (Qualters 2016). Although the LCME and HAPS have made efforts to include either a standard (LCME 2016) or a broader process learning goal (HAPS 2018) in their curricular guidelines to guide the incorporation of cultural competencies into instruction, the LCME standard is not specific enough to define all that cultural competence entails, and the HAPS learning goal is too broad to address cultural competencies fully. In fact, none of the organizations researched in this paper have really established specific learning objectives or standards for diversity and inclusion that can be used to educate students in the programs corresponding to these societal members’ institutions of higher learning. As a result, more effort needs to be invested by all professional societies with relevant anatomy and/or physiology content in the development and implementation of specific, measurable, attainable, realistic, and timely (SMART) diversity and inclusion goals or standards that will impact current and future learners. The policies, objectives, goals, or standards that are already possessed by some professional societies are mainly those used to drive the diversity and inclusion agenda among their membership, but not among the students that their members might teach.

Beyond the Classroom

Outside of the classroom, a set of diversity and inclusion goals or standards that guide departments, institutions, or organizations in nurturing a community of diverse individuals who feel included is also important. While a number of professional societies mentioned in this paper have adopted Diversity and Inclusion statements, founded Diversity and Inclusion committees, and/or published or posted Diversity and Inclusion resources, there is still much room for growth and improvement within these organizations in addition to those that lack diversity and inclusion content on their websites.

The movement to embrace diversity and inclusion should also take place on departmental and institutional levels. Although many institutions and departments have made such efforts, readers should contemplate the level of effort that their own departments and institutions have dedicated to promoting diversity and inclusion for their stakeholders. A group of authors under the editorship of Ferdman and Deane (2014) have published a textbook that describes frameworks for comprehending inclusion and presents both individual and organizational perspectives and practices of diversity and inclusion. Departmental, institutional, or organizational members can find a wealth of helpful ideas for transforming the way their respective entities contemplate and apply diversity and inclusion principles. The text also mentions global diversity and inclusion standards that can be referenced by any department, institution, or organization that wants to launch its own Diversity and Inclusion program and then utilize it to measure its progress toward achieving five levels of benchmarks into which the thirteen diversity and inclusion categories are organized (O’Mara and Richter 2016). These resources can be consulted by any group relevant to anatomy or anatomy and physiology that wants to start, evaluate, and/or improve its diversity and inclusion program. Additional materials can be found on the websites of those organizations with diversity and inclusion resources outlined in Appendix 1.

Diversity in Anatomy Today and Tomorrow

This section discusses the current status of this generation of students in general and the status in the field of anatomy, the potential directions toward which the field might expand in the future, the significance of cross-cultural interactions in the field, and the importance of professionals being well versed in the literature to provide the most relevant and practical learning experiences to students.

Teaching Tech-Savvy Learners

Students born in the twenty-first century or even the late twentieth century, such as current undergraduate and professional students, have been immersed in the World Wide Web, smart phones, social media, and high-resolution graphics. These students have been aptly named the ‘digital generation.’ In fact, several authors have dubbed
them “Digital Native” students whose ever-expanding technological demands must be met with newer computer-based innovations (Prenskey 2001, Palfrey and Gasser 2008). In today’s world of rapidly evolving technology, various commercial entities compete for the attention of students and occupational consumers alike. With so many competing modes of communication and entertainment, faculty in institutions of higher learning must continue to be creative in their approaches to gaining and maintaining student interest in curricular learning opportunities. In fact, administrators often pressure faculty to use state-of-the-art computer-based equipment to diversify their traditional teaching. But, instructors, including those in anatomy or anatomy and physiology must remember that technology is only a tool and that it is only useful if it improves students’ learning. Thus, educators should continuously monitor and reflect upon their instruction and make adjustments when necessary.

The Diversifying Landscape of Anatomical Research and Education
Traditionally, research in anatomy has involved “bench science” research whereby the investigator might discover a new anatomical structure or explore the common anatomical variants that exist for various structures. Now, anatomy is evolving and, in a way, diversifying published research to include educational scholarship, applications to technologically-enhanced procedures and skills, and novel approaches to clinical anatomy. These areas of professional growth and advancement allow instructors to bring both current and cutting-edge content and strategies into their anatomy or anatomy and physiology curricula. Then, by conducting studies in their own classrooms and institutions, educators can analyze the results of their students’ performance and share them with a diverse global anatomical community.

There are a number of emerging anatomy programs that emphasize educational research in the anatomical sciences. The Clinical Anatomy Program at UMMC is one such program that has been in existence for only five years. Additional anatomy education programs exist at the University of Colorado Anschutz Medical Campus, Indiana University, the University of Nebraska Medical Center, and the University of Western Ontario, just to name a few. A more complete list of graduate programs in anatomy can be found on the American Association for Anatomy’s website (Rowan 2018). At these institutions, the scope of educational research topics ranges from assessment, learning strategies, technological innovations, curricular design, and more. These topics, which have been explored in gross anatomy, developmental anatomy, neuroanatomy, and histology courses for health professional students have also been explored in human anatomy and physiology courses for undergraduate students. Researches in all of these disciplines have multiple opportunities each year to interact and network with one another at regional and annual meetings hosted by professional societies such as the American Association of Clinical Anatomists (AACA), the American Association for Anatomy (AAA), and the Human Anatomy and Physiology Society (HAPS).

Cross-Cultural Interactions in Anatomy
Today, advanced technology has established convenient communication via email, text messaging, social media, online blogs and open forums, teleconferences, and web meetings for people around the world. International anatomical societies have grown in popularity, and they provide platforms for researchers and educators from diverse groups to exchange ideas and discuss new findings and results. Such interactions allow attendees to establish cross-cultural collaborations that provide meaningful and unique insights into study designs and interpretations. Moreover, such studies that incorporate multiple global perspectives further enrich learning experiences and increase the robustness of studies. An excellent opportunity for in-person international, cross-cultural interactions occurs every four-to-five years when the professional anatomy society composed of the majority of the anatomical associations around the world, the International Federation of Associations of Anatomists (IFAA), holds its Congress. The 19th Congress was held this year in London, United Kingdom, August 9-11, 2019. The society includes organizational members from around the globe. According to the information posted on the IFAA website (IFAA History 2019), the organization is comprised of nearly 50 member societies, and it is steadily growing. In 2024, the 20th Congress of the IFAA, was approved by the IFAA delegates to be held in Gwangju, South Korea. In addition, the Human Anatomy and Physiology Society will hold its 2020 annual conference in Ottawa, Canada. Dr. Kevin Petti, a HAPS member and anatomy and physiology instructor at San Diego Miramar College, offers a summer course called Connecting Art and Anatomy through the HAPS Institute, a platform for HAPS members to earn professional development (HAPS Institute 2019). During this twelve-day experience, students visit six cities in Italy where they examine renderings of the human body in painting and sculpture. Furthermore, members of the International Association of Medical Science Educators (IAMSE) have an opportunity to attend the IAMSE Integration in Medical and Health Science Education symposium in Kuala Lumpur, Malaysia, in February 2020, preceding the Association for the Study of Medical Education (ASME) Ottawa Conference. The Association for Medical Education in Europe (AMEE) is another society through which members can attend other international conferences. The 2019 AMEE Conference will be held in Vienna, Austria, in August, and the 2020 AMEE Conference will be held in Glasgow, United Kingdom, in September.

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Not all professionals in anatomy, however, have to travel thousands of miles for multicultural experiences in the field. Exposure to diversity and inclusion could be as simple as a mentor-mentee relationship, especially if the mentor and mentee represent two distinct cultural backgrounds. In this particular case, the authors of this paper are both American citizens. Nevertheless, while the first author (mentor) was born and raised in the United States, the senior author (advisor) was born and raised in China. Since English is the senior author’s second language, this relationship has allowed the first author to assist in the review of grammar and spelling in manuscripts while the second author reviewed the scholarly content. This relationship has also allowed for cross-communication of the authors’ differing heritages between one another. However, mentor-mentee relationships in which either the mentor or mentee is still learning the language of the other can take on many levels of diversity given the variety of languages and cultures possessed by students and faculty in the field of anatomy or anatomy and physiology.

The Future of Anatomy Education
With the advent of the technological age, students in colleges and professional schools are very different from students over twenty years ago, and they will continue to change. Instructors, including those in anatomy and/or physiology must continue to adapt and diversify their teaching by incorporating evidence-based instructional practices and being well versed in the literature to provide the most relevant and practical learning experiences to students, especially if they intend to be educators or healthcare providers in the future. The future success of anatomy as a field and of the anatomist or anatomy educator as a profession depends on this flexibility and response to ever-changing technology, knowledge, and student and faculty needs.

Conclusions
Overall, the push for institutional policies for the promotion of diversity and inclusion both nationally and worldwide has been a positive endeavor that has improved and increased interactions between individuals of varying characteristics. Diversity and inclusion activities have the potential to foster such interactions in anatomy and anatomy and physiology classrooms. Several professional societies including anatomy or anatomy and physiology educators have made great strides in providing diversity and inclusion content on their websites, but there is much room for improvement in terms of these and other organizations’ contributions to diversity and inclusion. The dwindling supply and growing demand for anatomy educators coupled with the current limited diversity in institutions of higher learning further amplifies the need for more diverse and inclusive anatomy education programs. Future efforts in diversity and inclusion have the potential to foster a more culturally sensitive and engaged community of anatomy and/or physiology educators as well as a global society of citizens who celebrate diversity and embrace each other in inclusive environments.

About the Authors
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Literature cited


American College of Physicians (ACP). 2019. [accessed in 2019 Jul 17]. Available from: https://www.acponline.org/?&gclid=EAIaIQobChMI76-uJNDE4wIvbf_jBx38BQgFEAAWASAAElbqPD_BwE&cpid=8a8ae4b331b3b50a0131be21d3370017&jsf=108d109e-8f33-4756-9c08-c348dbd7af32-33443


Diversity and Inclusion in Anatomy and Physiology Education, Degree Programs, and Professional Societies


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Diversity and Inclusion in Anatomy and Physiology Education, Degree Programs, and Professional Societies


Diversity and Inclusion in Anatomy and Physiology Education, Degree Programs, and Professional Societies


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Appendix 1. The Status of Diversity and Inclusion Content on the Websites of Professional Societies and Organizations That Include Anatomists or Anatomy Educators.

<table>
<thead>
<tr>
<th>Category or Classification</th>
<th>Organization or Professional Society</th>
<th>D &amp; I Standards, Policies, or Objectives</th>
<th>D &amp; I Mission or Vision Statement</th>
<th>D &amp; I Committee or Task Force</th>
<th>Webpage or Website with Resources</th>
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<tbody>
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<td>Anatomy- and/or Physiology-Specific Professional Societies</td>
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<td>In Progress</td>
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<td>No</td>
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</tr>
<tr>
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<tr>
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<td></td>
<td>American Society of Radiologic Technologists (ASRT)</td>
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<td></td>
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<td>Council**</td>
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<th>Category or Classification</th>
<th>Organization or Professional Society</th>
<th>D &amp; I Standards, Policies, or Objectives</th>
<th>D &amp; I Mission or Vision Statement</th>
<th>D &amp; I Committee or Task Force</th>
<th>Webpage or Website with Resources</th>
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<tr>
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<tr>
<td>Medicine</td>
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<td>No</td>
<td>Council and Committee</td>
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</tr>
<tr>
<td></td>
<td>American College of Physicians (ACP)**</td>
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<td>Sub-committee</td>
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<td>Group**</td>
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<td>Committee</td>
<td>Resources (but no dedicated page)</td>
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<tr>
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<td>American Nurses Association (ANA)</td>
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<td>Pharmacy</td>
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<td></td>
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<td></td>
<td>American Pharmacists Association (APA)</td>
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<td>Category or Classification</td>
<td>Organization or Professional Society</td>
<td>D &amp; I Standards, Policies, or Objectives</td>
<td>D &amp; I Mission or Vision Statement</td>
<td>D &amp; I Committee or Task Force</td>
<td>Webpage or Website with Resources</td>
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<td>---------------------------------</td>
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<tr>
<td>Health Professional Societies with Content Integrated into Anatomy Curricula</td>
<td>American Society of Clinical Pathology (ASCP)*</td>
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<tr>
<td></td>
<td>College of American Pathologists (CAP)</td>
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<td></td>
<td>United States and Canadian Academy of Pathology (USCAP)</td>
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<td></td>
<td>Radiological Society of North America (RSNA)</td>
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<td>Mixed Basic and Clinical Science Professional Societies</td>
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<td></td>
<td>Federation of American Societies for Experimental Biology (FASEB)*</td>
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<td></td>
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<tr>
<td></td>
<td>International Association of Medical Science Educators (IAMSE)</td>
<td>No</td>
<td>No</td>
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### Diversity and Inclusion in Anatomy and Physiology Education, Degree Programs, and Professional Societies

<table>
<thead>
<tr>
<th>Category or Classification</th>
<th>Organization or Professional Society</th>
<th>D &amp; I Standards, Policies, or Objectives</th>
<th>D &amp; I Mission or Vision Statement</th>
<th>D &amp; I Committee or Task Force</th>
<th>Webpage or Website with Resources</th>
</tr>
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<tr>
<td>Other Anatomy-Relevant Professional Societies</td>
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<td>No**</td>
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<tr>
<td></td>
<td>American College of Veterinary Surgeons (ACVS)</td>
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<tr>
<td></td>
<td>American Society of Animal Science (ASAS)</td>
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<td></td>
<td>American Society of Embalmers (ASE)</td>
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<td>American Veterinary Medical Association (AVMA)</td>
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<td>Paleontological Society (PS)</td>
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<td>Yes</td>
<td>Yes</td>
<td>No</td>
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<tr>
<td></td>
<td>Society for Medical Anthropology (SMA)^</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
</tbody>
</table>

This table does not represent an exhaustive list of all of the professional societies and organizations that might contain anatomists, anatomy educators, anatomy and physiology educators. This table also does not include many of the professional societies representing specialties or subspecialties within the various healthcare disciplines. In addition, this table mainly features national and international organizations whose headquarters are located in the United States, and while it does include a few organizations in Europe, it in no way represents all of the anatomical organizations within other countries nor does it represent all of the foreign organizations that might contain anatomists, anatomy educators, or anatomy and physiology educators. The citations for the website homepages of all the organizations included in this table are provided within the list of references in this paper.

^Note: While the American Physiological Society (APS) does not have a committee, task force, or interest group devoted to diversity and inclusion specifically, it does have a committee devoted to women in physiology and another devoted to minority affairs. In addition, the organization has an interest group including members engaged in sex and gender research in the field.
Note: Although the Human Anatomy and Physiology Society (HAPS) does not have official standards, policies, or objectives governing or guiding the implementation of diversity and inclusion activities or content delivery, it does include broader process goals beyond its learning goals posted on its website (HAPS 2018). These goals are broad enough to allow instructors flexibility to incorporate their own diversity and inclusion objectives in their classrooms.

††Note: The International Federation of Associations of Anatomists (IFAA) is not an organization composed of individual members, but rather an organization composed of different anatomical professional societies throughout the world. It holds its congress once every five years. However, the organization does have a website and an officer team. For the sake of brevity, the anatomical associations in countries other that the United States were not included in this table. However, a list of these societies along with links to their respective websites can be found by visiting the IFAA website (IFAA Societies 2019). A number of these individual societies may post information pertaining to diversity and inclusion on their websites. While the IFAA does not have a Diversity and Inclusion Committee, it does have a Federative International Committee for Equality and Diversity in Anatomy (FICEDA).

*Note: The Association of Schools and Colleges of Optometry (ASCO) has a Diversity and Cultural Competency Committee (DCCC).

**Note: While the Physician Assistant Education Association (PAEA) does not have a committee of task force dedicated to diversity and inclusion, it does have a council dedicated to both endeavors. In addition, the organization has a Cultural Competencies Review Committee which is responsible for ensuring the promotion of cultural sensitivity among its members.

***Note: There are a number of specialties within the field of dentistry, such as orthodontics and periodontics, but for the sake of brevity, the professional societies representing these specialties were not included in this table. Please feel free to search the web for a list of these additional organizations and their respective websites.

‡Note: Not included in this table is the American College of Surgeons (ACS). There are many organizations devoted to various surgical specialties in which anatomy is highly relevant; however, for the sake of brevity, these professional societies were not included in this table. Feel free to visit the website of the American College of Surgeons (ACS) to find a list of these additional organizations (ACS Other Surgical Societies of Interest 2019). This list may or may not be exhaustive.

‡‡Note: The American Association of Colleges of Nursing (AACN) has a Diversity, Equity, and Inclusion Group (DEIG) in place of a committee.

‡‡‡Note: While the American Nurses Association (ANA) does not have an official set of standards or policies, vision or mission statement, or committee dedicated to diversity and inclusion, it does have a webpage dedicated to Diversity Awareness on its website. Although this webpage is not dedicated to diversity and inclusion specifically, it does include resources pertaining to topics of diversity.

● Note: Although the American Society of Clinical Pathology (ASCP) does not include any of the content mentioned in the table on its website, it does host a Leadership Institute with an associated Certificate Program through which diversity and inclusion are one of the areas of leadership featured in the syllabus (ASCP Leadership Institute 2019).

●● Note: The American College of Radiology (ACR) does not have a committee specifically devoted to diversity and inclusion, but it does have a Commission for Women and Diversity which has a set of goals, a statement on diversity and inclusion, and a webpage with resources. The commission also publishes an annual report titled “Diversity Report: Excellence Through Diversity and Inclusion” (ACR 2019).

●●● Note: While the American Association for the Advancement of Science (AAAS) does not have policies, statements, committees, or a webpage with resources specifically devoted to diversity and inclusion, the organization in conjunction with the Association of American Universities (AAU) did release a comprehensive guide titled “Handbook on Diversity and the Law: Navigating a Complex Landscape to Foster Greater Faculty and Student Diversity in Higher Education” for establishing legally sustainable diversity programs for campuses in 2010 (Ham 2010).
Note: The Federation of American Societies for Experimental Biology (FASEB) is not composed of individual members, but rather multiple biological associations, such as the American Association for Anatomy (AAA). FASEB has a webpage devoted to its Diversity Resources Program. “The FASEB Diversity Resources Program is supported by grants from the National Institute of General Medical Sciences of the National Institutes of Health: Maximizing Access to Research Careers (MARC) Ancillary Training Program (T36) and Innovative Programs to Enhance Research Training (IPERT) (R25). [...] The FASEB Diversity Resources for Enrichment, Access and Mentoring (DREAM) activities (supported by MARC and IPERT grants) focus on skill development courses/workshops, structured mentoring activities and outreach programs which are designed to help create a highly skilled and diverse biomedical research workforce from all groups” (FASEB 2017).

Note: Although the American Anthropological Association (AAA) does not have a committee specifically devoted to diversity and inclusion, it does have a committee known as the Members’ Programmatic, Advisory and Advocacy Committee (MPAAC) which focuses on issues of diversity and inclusion such as human rights, labor and workforce, minority issues, and gender equity.

Note: While the Palaeontological Association (PA) does not have content pertaining to diversity and inclusion on its website, the organization did hire a British company known as Parigen Limited to conduct a diversity study on the organization in 2018. This study (Council Diversity Group 2018) can be found on the organization’s website homepage.

Note: The Society for Medical Anthropology (SMA) is a section of the American Anthropological Association (AAA), but it does have its own website (SMA 2017).
Infantile Spasms: The Role of Prenatal Stress and Altered GABA Signaling

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Abstract
Infantile spasms (IS) is a rare epileptic disorder occurring in children under the age of one that can often lead to severe developmental delays throughout life. Though over 200 etiologies have been associated with this disorder, many cases remain unexplained. Research into the etiology of IS has implicated causes such as exposure to prenatal stress or changes in GABAergic signaling. Here, we describe recent findings that draw a direct connection between prenatal stress, altered GABA signaling, and the development of IS. We also discuss how these findings can be used in the classroom to enrich discussions of nervous system physiology, development, and disease. https://doi.org/110.21692/haps.2019.017

Key words: neurophysiology, infantile spasm, epilepsy, GABA, maternal stress

Introduction
Infantile spasms (IS) is a disorder characterized by infantile epileptic spasms. IS is estimated to occur in about 2.5 – 5.0 in 10,000 live births (Paciorkowski et al. 2011; Shi et al. 2012). There are three defining features used in the diagnosis of IS. The first is the occurrence of repetitive spasms, with each spasm consisting of a short flexion, a short contraction, or both, for one to two seconds (Swann and Moshe 2012). The second defining feature is that spasm onset occurs in children one year of age or younger, most typically between three to seven months of age (Swann and Moshe 2012; Pavone et al. 2014). The third defining feature, which sets IS apart from other forms of infantile epilepsies, is hypsarrhythmia (Pavone et al. 2014). Hypsarrhythmia is an abnormal brain pattern on an electroencephalogram (EEG), characterized by chaotic, high-voltage, and slow waves along with asynchronous waves between each hemisphere and within each hemisphere of the cortex (Shields 2006). Several subtypes of IS have been recognized based on the particular set of symptoms present. The most common of these, West Syndrome, is defined by the appearance of all three characteristics of IS accompanied by developmental delays that become apparent as the child grows (Pavone et al. 2014).

Although the spasms associated with IS typically resolve by age five, the consequences of those early epileptic events can have repercussions throughout a patient’s lifetime. IS is associated with serious developmental delays. Only seventeen percent of those affected by IS will have an IQ greater than 85, and only 15 – 25% of patients achieve a developmental outcome that would be characterized as normal (Nelson 2015). The most common developmental delays include autism, which occurs in about 15 – 33% of cases, and intellectual disability, present in about 45% of cases (Shields 2006). Since the spasms occur during a critical period of development, adverse developmental outcomes can only be prevented when treatments yield complete control of the spasms (Shields 2006). Unfortunately, there are only two currently available treatment options; neither guarantees complete spasm control, and both have significant drawbacks (Shields 2006). The first treatment option, adrenocorticotropic hormone (ACTH), has been shown to reduce spasm activity by 42 – 87% within two weeks of treatment. However, patients taking ACTH commonly experience relapse along with serious side effects such as electrolyte imbalances, delays in growth, suppression of the immune system, and cardiomyopathy (Nelson 2015). A second treatment option, the anticonvulsant drug Vigabatrin, is preferred because it has fewer side effects (Shields 2006). However, this drug also has significant limitations, including a 16 – 21% relapse rate and visual impairments in 15 – 30% of cases (Nelson 2015).

Because IS is associated with serious lifelong developmental delays there is significant interest in identifying its underlying causes, with the hope that increased understanding will aid in the development of more effective treatment approaches and preventative measures. Previous research has identified multiple conditions associated with IS, including tuberous sclerosis complex, Down syndrome, cortical malformations, and prenatal infections (Shi et al. 2012; Swann and Moshe 2012; Yuskaitis et al. 2018). However, about a third of patients are diagnosed with “cryptogenic” IS, in which no underlying cause can be identified (Shields 2006; Yuskaitis et al. 2018). There is an urgent need to elucidate the cause(s) of cryptogenic IS, as there is some evidence to suggest that IS etiology can influence treatment efficacy (Garcia-Penas and Jimenez-Legido 2017; Liang et al. 2017; Roldan 2017; Ko et al. 2018).
A number of potential mechanisms have been proposed for cryptogenic IS. Some studies have implicated altered gamma-aminobutyric acid (GABA) signaling, because multiple patients suffering from IS have been found to be carrying mutations in GABA pathway genes (Galanopoulou 2010; Edvardson et al. 2013; Kelsom and Lu 2013; Olivetti et al. 2014; Papandreou et al. 2016). Other studies, using animal models of IS, suggest that exposure to prenatal stress might contribute to the development of IS, although the precise mechanism of action is unknown (Shi et al. 2012; Yum et al. 2012). Although these two proposed etiologies – altered GABA signaling and exposure to prenatal stress – have been studied independently, more recent research suggests they may actually be directly linked. In this review, we will describe recent findings suggesting that prenatal stress causes alterations in the GABA pathway, thereby contributing to the development of IS. We will also discuss how these findings can be used in the classroom to enrich discussions of nervous system physiology, development, and disease.

Role of the GABA pathway during normal nervous system development

GABA is a neurotransmitter that acts as an important inhibitory signal in the adult central nervous system (Li and Xu 2008). GABAergic neurons synthesize GABA from the amino acid glutamate in a reaction that is catalyzed by glutamic acid decarboxylase (GAD). When GABA is released from GABAergic neurons, it binds to receptor proteins on postsynaptic neurons (Li and Xu 2008; Wang and Kriegstein 2009). GABA’s principal receptors are the GABA_A receptors. GABA_A receptors are ionotropic; when bound to GABA, GABA_A receptors trigger the opening of Cl^- channels (Li and Xu 2008; Wang and Kriegstein 2009). In the mature brain, most cells express KCC2 (K^-Cl^- cotransporter, which pump Cl^- ions out of the cell, thus establishing a lower intracellular concentration of Cl^- (Wang and Kriegstein 2009). As a result, when GABA binds to GABA_A receptors, Cl^- ions flow into the postsynaptic neuron, hyperpolarizing the neuron and leading to an inhibitory effect (Li and Xu 2008; Wang and Kriegstein 2009).

While GABA is well established as an inhibitory signal throughout adulthood, its role differs during development of the nervous system. During prenatal development and into the first two weeks of infancy, most neurons express the NKCC1 cotransporter rather than the KCC2 cotransporter (Kirmse et al. 2018). NKCC1 (a Na^-K^-Cl^- cotransporter) pumps Cl^- ions into the neuron, increasing the intracellular concentration (Li and Xu 2008; Wang and Kriegstein 2009). Therefore, when GABA binds to GABA_A receptors in immature neurons, Cl^- ions flow out of the postsynaptic neuron, depolarizing the neuron and leading to an excitatory effect (Li and Xu 2008; Wang and Kriegstein 2009; Kirmse et al. 2018).

The excitatory function of GABA is essential for many aspects of central nervous system development (Wang and Kriegstein 2009; Kirmse et al. 2018). For example, GABA directs the proliferation and migration of distinct populations of progenitor cells (Wang and Kriegstein 2009; Wu and Sun 2014). In addition, because most newly formed neurons express GABA receptors before glutamate receptors, excitatory GABA is critical in the early formation of neural networks (Wang and Kriegstein 2009). However, as the brain develops, GABA transitions from an excitatory function to the inhibitory role it will play throughout most of childhood and adulthood. This excitatory-to-inhibitory switch is driven by changes in gene expression - increased expression of KCC2 and decreased expression of NKCC1 – and normally occurs in the second week of the postnatal period (Kirmse et al. 2018). Since GABA signaling is such a prominent and critical driving force in the formation of neural circuits in the central nervous system during development, changes in prenatal and postnatal GABAergic signaling would be expected to have wide-ranging and long-lasting effects on the central nervous system.

Maternal stress can affect prenatal development

In humans, exposure to stress (both physical and emotional) stimulates the hypothalamic-pituitary-adrenal (HPA) axis, causing a variety of responses (Chrousos 2008; Brunton 2013; Goldstein et al. 2013). Part of the stress response includes the release of stress hormones, such as corticotrophin releasing hormone (CRH), catecholamines such as epinephrine, and glucocorticoids such as cortisol (Chrousos 2008; Goldstein et al. 2013). These stress hormones act on multiple body systems, including the central nervous system, cardiovascular system, and immune system, to induce a wide range of immediate effects on the body (Chrousos 2008; Goldstein et al. 2013). While the stress response is an adaptive trait, acute and chronic stress are also thought to have many adverse effects and are linked to diseases such as hypertension and metabolic disorders in adults (Chrousos 2008).

Pregnant women undergo the same physiological responses to stress, and there is evidence to suggest that the resulting hormones, particularly CRH and glucocorticoids such as cortisol, may adversely affect the developing fetus (Chrousos 2008). Although an enzyme called 11β-hydroxysteroid dehydrogenase (11βHSD2) is expressed in the placenta to prevent glucocorticoids from crossing the placental barrier, maternal stress can suppress the expression of 11βHSD2 (Huang 2014). As a result, when a pregnant woman experiences acute or chronic stress, glucocorticoids are able to cross the placental membrane and reach the fetal body. Once glucocorticoids enter the fetal body, they can potentially affect development of multiple organs and organ systems, including the central nervous system (Chrousos 2008).
Because of the pivotal processes taking place during embryonic development, the developing fetus is especially vulnerable to changes in its environment (Sandman et al. 2011). In the central nervous system, crucial events such as neurogenesis, differentiation and proliferation of neuronal progenitor cells, synaptic formation, and the emergence of the neural circuit occur during prenatal development (Shi et al. 2012). Exposure of the fetus to gestational stress can specifically impact central nervous system development (Sandman et al. 2011). Prenatal exposure to excess glucocorticoids is thought to alter gene expression in the brain both via changes in transcription factor activity and via epigenetic modifications, such as DNA methylation and histone modifications (Fine et al. 2014; Whirledge and Cidlowski 2010). Fetal exposure to excess glucocorticoids can also interfere with neural signaling in the fetal brain, including the GABA pathway (Iacobas et al. 2013). These alterations in the developing central nervous system are hypothesized to have impacts that persist well beyond birth, increasing the risks for disorders such as anxiety, autism, schizophrenia, ADHD, and IS (Wang et al. 2017; Negrón-Oyarzo et al. 2016; Sandman et al. 2011). Indeed, a number of recent studies have shown that prenatal stress specifically alters components of the GABAergic pathway and increases susceptibility to IS in particular (Shang et al. 2010; Stevens et al. 2013; Uchida et al. 2014; Baek et al. 2016; Shi et al. 2016; Kwon et al. 2018; Vangeel et al. 2017).

Effects of prenatal stress on the GABA pathway and spasm susceptibility

A number of recent studies have shown that prenatal stress can directly alter GABAergic signaling in the fetal and neonatal brain (Stevens et al. 2013; Uchida et al. 2013; Baek et al. 2016; Shi et al. 2016; Kwon et al. 2018; Vangeel et al. 2017). These changes occur at multiple levels of the GABA pathway. Furthermore, several of these studies directly link prenatal stress and changes to GABAergic signaling with increased risk of IS (Shang et al. 2010; Baek et al. 2016; Shi et al. 2016; Kwon et al. 2018).

Prenatal stress can affect GABAergic signaling in multiple ways. First, prenatal stress has been shown to reduce the proliferation of GABAergic neurons during embryonic development (Stevens et al. 2013; Uchida et al. 2014). Prenatal stress also impairs the migration of GABAergic neurons during embryonic development, at least in part due to decreased expression of genes required for migration (Stevens et al. 2013). As a result, there is a general decrease in the number of GABAergic neurons present in the neonatal cortex of prenatally stressed offspring (Stevens et al. 2013; Uchida et al. 2014; Baek et al. 2016; Kwon et al. 2018). Prenatal stress also affects downstream components of the GABA pathway during embryonic development. A gene encoding one of the GABA receptors has been found to undergo increased DNA methylation following exposure to prenatal stress, and GABA receptor binding function is also reduced following exposure to prenatal stress (Vangeel et al. 2017; Baek et al. 2016). Collectively, these changes could significantly impair GABAergic signaling during embryonic development in prenatally stressed offspring.

In addition to changes in GABAergic neurons and GABA receptors during embryonic development, the excitatory-to-inhibitory switch that normally occurs in the neonatal period may also be affected by prenatal stress. The excitatory-to-inhibitory switch is normally driven by downregulation of NKCC1 and simultaneous upregulation of KCC2 shortly after birth (Kirmse et al. 2018). However, KCC2 expression is significantly reduced in the neonatal cortices of prenatally stressed offspring (Baek et al. 2016; Kwon et al. 2018). Thus exposure to prenatal stress might cause a delay in the excitatory-to-inhibitory switch.

These recent studies have shown that prenatal stress impairs the excitatory GABAergic signaling that occurs during embryonic development, which could potentially disrupt development of the central nervous system (Stevens et al. 2013; Uchida et al. 2014; Baek et al. 2016; Vangeel et al. 2017; Kwon et al. 2018). In addition, prenatal stress disrupts the postnatal excitatory to inhibitory switch, which could impair inhibitory GABA signaling in the neonatal brain (Baek et al. 2016; Kwon et al. 2018). These pre- and post-natal changes to GABAergic signaling could each contribute to increased IS susceptibility. Indeed, prenatal stress increases likelihood and severity of infantile spasms in rodent models of IS (Baek et al. 2016; Shi et al. 2016; Kwon et al. 2018). Human studies have also found a correlation between prenatal stress and IS (Shang et al. 2010). While future studies are needed to determine the mechanism by which prenatal stress directly alters components of the GABAergic pathway, these studies build a compelling argument supporting a model in which prenatal stress directly alters GABAergic signaling in the developing central nervous system, leading to increased susceptibility to IS.
Infantile Spasms: The Role of Prenatal Stress and Altered GABA Signaling

Conclusion
Multiple recent studies demonstrate that prenatal stress and associated changes in GABAergic pathways may present an important mechanism increasing susceptibility to the development of IS during infancy. These findings are clinically relevant as they may help in the development of more effective prevention and treatment plans for IS. Future studies should continue to elucidate the mechanisms by which prenatal stress alters GABA signaling and develop drugs that target GABA pathway components for the treatment of IS.

Classroom Implementation Guide
Because GABA serves as the primary inhibitory signal in the adult nervous system, GABAergic signaling is an important topic covered in nearly all physiology courses. This manuscript can be used to reinforce basic concepts of GABAergic signaling, incorporate current research in the field of neurophysiology into the classroom, and encourage students to discuss clinical applications of the GABA pathway. Here is a suggested outline for use of this manuscript in a physiology classroom:

1. Introduce the basic concepts of GABAergic signaling.
2. Ask students to discuss how changes in the GABA pathway might result in epilepsy. The discussion should lead to the conclusion that a decrease in GABA function could cause nervous system disinhibition or inappropriate excitation, inducing seizure.
3. Ask students to read the section of this manuscript that discusses the role of the GABA pathway during development, and use this information to predict how a delay in the excitatory-to-inhibitory switch might affect infants. Students should conclude that the delay might cause seizures.
4. More advanced students can read the rest of the paper to learn about the role of prenatal stress in altering GABAergic signaling and causing IS. Following this reading, students can be asked to discuss how these recent findings should influence maternal care guidelines and research into more effective treatments for IS. These discussions will reinforce student understanding of GABAergic signaling throughout both development and adulthood and encourage them to apply their knowledge to clinical and therapeutic applications.

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The Effects of Ketogenic Diets on Psychiatric Disorders Involving Mitochondrial Dysfunction: A Literature Review of the Influence of Dieting on Autism, Depression, Anxiety, and Schizophrenia

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Abstract
Mitochondrial dysfunction has been linked to many psychiatric disorders. Ketogenic diets have been shown to reduce mitochondrial dysfunction and thus may be helpful to patients who suffer from these disorders. In this article, we review the effects of a ketogenic diet in patients with psychiatric disorders such as autism, depression, anxiety, and schizophrenia. Mitochondrial dysfunction, and its reversal by a ketogenic diet resulting in the relief of mental disorders, would be an excellent teaching topic on the consequences of altered cellular biology and a discussion of mitochondrial injury as one of the causes of cellular necrosis. Similarly, this information will be important in a discussion of how diet might be beneficial in the treatment of certain mental disorders. https://doi.org/10.21692/haps.2019.002

Key words: ketogenic diet, mitochondria, psychiatric conditions

Introduction
Mitochondrial phosphorylation dysfunction is associated with many psychiatric disorders including autism, depression, anxiety, and schizophrenia. A switch from a regular diet to a ketogenic diet, which induces ketosis, a high level of ketone bodies in the blood, has been shown to minimize mitochondrial dysfunction and thus may help to improve symptoms of some psychiatric disorders. There are several mechanisms by which ketogenic diets appear to improve mitochondrial dysfunction including the release of antioxidants, the reduction of inflammation, and the reduction of oxidative stress. In this article, we review the effects of a ketogenic diet in patients with specific psychiatric disorders.

Mitochondrial dysfunction, and its reversal by a ketogenic diet resulting in the relief of mental disorders, will serve as an excellent clinical exemplar in the discussion of the consequences of alterations in cellular and molecular biology, the applications of nutrition science, and topics in pathophysiology. For example, a discussion of the neurotransmitter dopamine might lead to a discussion of how Parkinson’s disease is the result of the loss of dopaminergic neurons due to mitochondrial dysfunction. The effect of mitochondrial dysfunction in psychiatric disorders, as presented here, reinforces the relationship between alterations in cellular structures and resulting abnormalities of function.

Diet is one approach to fighting obesity. Low-carbohydrate, high-fat diets, including their intense version, the ketogenic diet, have become popular dietary regimes for several reasons. Ketogenic diets have been acclaimed as an effective method to control body weight and blood glucose levels (Azar et al. 2016). Recent studies further report that this regimen has positive effects on the central nervous system (CNS) (Maalouf et al. 2009; Mattson et al. 2018). Ketogenic diets have produced beneficial effects in multiple psychiatric disorders such as autism, depression, anxiety, and schizophrenia (Evangeliou et al. 2003; Herbert and Buckley 2013; Kashiwaya et al. 2013; Mantis et al. 2009; Palmer 2017; Sussman et al. 2014; Wlodarczyk et al. 2018). There could be several reasons for the reported improvements. For example, the ketogenic diet has been shown to reduce inflammation and oxidative stress (Johnson et al. 2007). The associated metabolic switch in the cellular fuel source is accompanied by cellular and molecular adaptations of neural networks in the brain that enhance their functionality and bolster their resistance to stress, injury, and disease (Mattson et al. 2018). Further understanding of the mechanism of action of the ketogenic diet suggests that ketosis, a higher than normal level of circulating ketone bodies, induced by a ketogenic diet, helps reverse the mitochondrial dysfunction commonly noted in many psychiatric disorders. This article will examine the beneficial effects of ketogenic diets on these disorders, and attempt to explain common theories as to why these positive interactions occur.
The Role of Mitochondrial Defects in Psychiatric Disorders

Mitochondria are the most prominent source of ATP in the cell. Approximately 98% of cellular ATP is generated by these intracellular membranous structures. In addition to energy production, mitochondria are important in a variety of functions ranging from calcium buffering to regulation of apoptosis (Fattal et al. 2006).

Calcium ion concentration in the cell is critical since high levels of calcium can trigger signal transduction cascades resulting in abnormal outcomes. Mitochondria help maintain calcium homeostasis by having the ability to accumulate calcium ions in an energy dependent manner and release calcium back into the cytoplasm. Extracellular cues resulting in the activation of the inositol triphosphate cell signaling cascade for example, increases intracellular calcium levels, which in turn increase mitochondrial calcium uptake. Greater than normal calcium uptake by the mitochondria may lead to increased metabolism and energy production or initiating apoptosis (Pinton et al. 2010; Santo-Domingo and Demaurex 2010).

In addition to calcium signaling and regulation of cell death, many chemical reactions that occur in the mitochondria result in the production of free radicals, nitric oxide, and hydrogen peroxide, which play a significant role in signaling and determining if apoptosis or proliferation is more appropriate in a given situation (Cadenas 2004). Other metabolites that are associated with the TCA cycle include acetyl coenzyme A, citrate, isocitrate, 2-oxy succinyl coenzyme A, succinate, fumarate, malate, and oxaloacetate (Frezza 2017).

The brain requires 25% of the body’s energy supply and a single neuron can consume 4.7 billion ATP molecules per second (Kramer and Bressan 2017). Given the enormous energy demand by the brain, continuous mitochondrial production of ATP may result in the build up of harmful agents such as reactive oxygen species (ROS). Higher ROS levels are shown to trigger a positive feedback mechanism resulting in an elevated production of ROS (Zorov et al. 2014) and thus, induce mitochondrial dysfunction. Mitochondrial dysfunction can be an influential factor in the development of psychiatric disorders. In fact, several types of mitochondrial dysfunction are being linked to autism, depression, anxiety, and schizophrenia (Allen et al. 2018; Griffiths and Levy 2017). Normal physiological processes, such as neurogenesis and the modulation of synaptic connections known as synaptic plasticity, demand large amounts of energy. Mitochondria on the dendrites and axons are responsible for calcium signaling, generating action potentials, development of new synapses, and remodeling mature synapses, all of which are integral to plasticity. During the formation of neuroplasticity, the OXPHOS must continuously produce ATP to meet high energy requirements. (Bergman and Ben-Shachar 2016).

The Ketogenic Diet and Mitochondrial Dysfunction

Due to the lack of availability of glucose associated with a ketogenic diet, the body switches to produce a large amount of ketone bodies, which in turn serve as alternate sources of energy in the metabolically active areas, such as the brain. The presence of ketone bodies, in place of glucose, appears to aid in repairing some of the damage observed in the psychiatric disorders simply by promoting mitochondrial reproduction and increasing energy production (Kramer and Bressan 2017).

Ketone bodies have also been known to possess neuroprotective properties (Cunnane et al. 2016). For example, ketogenic diets have been in clinical use for over a century as an anticonvulsive therapy. Neuroprotective qualities consist of the reduction of neuronal apoptosis and brain edema, and the production of increased levels of neurotrophins (Maalouf et al. 2009).

Though the exact mechanism of how ketogenic diets reverse some of the effects of mitochondrial dysfunction is unknown, there are several theories to explain the process, most of which are associated with the efficiency in the availability of ATP for normal neuronal function (Maalouf et al. 2009; Wlodarczyk et al. 2018). For example, ketogenic diets are thought to decrease the production of ROS and increase ATP and phosphocreatine levels, thus boosting metabolic efficiency. Another possibility is that ketogenic diets may exert a positive effect by limiting apoptosis as well as neuronal excitability (Maalouf et al. 2009). Another theory involves correction of the OXPHOS system. When there is mitochondrial damage and the OXPHOS system is not performing optimally, oxidative damage occurs to proteins, lipids, and DNA in the brain. Any
diet that has caloric restriction, which includes ketogenic diets, produces antioxidant properties and helps delay the damage caused by the oxidative effects (Maalouf et al. 2009). The final theory relates specifically to the ketogenic diet on schizophrenia in which Wlodziak et al. (2018) hypothesize that ketogenic diets change the ratio of gamma aminobutyric acid (GABA) and glutamate in such a way that there is an increase in the synthesis of GABA as well as glutamate metabolism. Ketogenic diets seem to compensate for the disrupted GABA levels in a schizophrenic brain, leading to possible improvement of symptoms of the disease (Wlodziak et al. 2018). Despite the gaps in our current knowledge about the exact mechanism, available evidence suggests that ketogenic diets are associated with symptomatic improvements in several psychiatric conditions.

The Ketogenic Diet and Autism
Autism is a neurodevelopmental disability that is characterized by deficits in communication and social interactions, as well as stereotypical behavior. The severity of this disability is characterized on a spectrum ranging from mild to severe. Diagnosis is determined using the Childhood Autism Rating Scale (CARS) which ranks symptoms resulting in the following scores: 15-29.5 as non-autistic, 30-36.5 as mild to moderately autistic, and 37-60 as moderate to severely autistic. The following symptoms are considered for classification: relating to people, imitation, emotional response, object use, body use, adaptation to change, visual response, listening response, taste, smell, touch response and use, fear/nervousness, verbal and nonverbal communication, activity level, and intelligent response consistency, and general impressions (Al Backer 2016).

Zarnowska et al. (2018) reported a case involving a six-year-old male with autism that revealed positive effects of a ketogenic diet. At the beginning of the study, the boy scored a 43 on the CARS, which indicates severe autism. He had trouble with emotional responses and social interactions, and he obsessively asked the same questions repeatedly. He scored an 82 on the Wechsler Intelligence Scale (WISC) IQ scale for children, which is considered borderline intellectually disabled. A ketogenic regimen was initiated at age six years and one month. Improvements became evident after one month on the diet. The boy was less aggressive and less hyperactive. After 16 months of being on the diet, another psychological evaluation was performed at age seven year and five months. This evaluation revealed a score of 27 on the CARS, which was a 16-point improvement. The improvements were listed as: less hyperactivity, fear, anxiety, emotional, and abnormal visual/auditory reactions, coupled with increased attention, improved use of objects, improved adaptability to change, and improved communication abilities. His WISC IQ tested at a 99, which is considered average (Zarnowska et al. 2018).

Similar findings were reported in another case study by Herbert and Buckley (2013). In this study, a female child started exhibiting autistic characteristics at four years of age. These included escalating tantrums, decreased eye contact, lack of social awareness or interest, and increased sensory hypersensitivities. Her language had regressed from an average four-year-old level to an 18-month level. At 11.5 years of age, the child started to experience grand mal seizures, prompting doctors to initiate a gluten/casein-free ketogenic diet at age 12. Along with improved seizure control, the child also experienced improved language and cognitive function, as well as improved social skills and increased calmness. Her initial CARS score was a 49 (severely autistic) and was reduced to a 17 (non-autistic) over the years (Herbert and Buckley 2013).

A pilot study by Evangeliou et al. (2003) involved 30 children, two were classified as mild to moderately autistic and the other 28 were classified as severely autistic. Results from this study provided additional evidence of the beneficial effects of ketogenic diets for autistic children. Eighteen of the 30 children completed six months of a ketogenic diet. Two children improved their CARS score by over 12 points. Eight children improved their score by an average of eight to twelve points. The remaining eight children achieved minor improvements of two to eight points (Evangeliou et al. 2003).

A study performed by Mantis et al. (2009) details the use of a ketogenic diet in male Mecp2(308/y) mice in reference to Rett Syndrome (RTT), an X-linked autistic spectrum neurological disorder. The disorder is characterized by impaired energy metabolism, motor impairment, social behavioral regression, and seizure susceptibility. Girls with RTT do not exhibit developmental problems until approximately 18 months of age. At this time, speech and behavioral regression become evident. Symptoms include hand wringing, anxiety, mental retardation, seizure, and other behaviors commonly associated with autism. After 30 days on a ketogenic diet, motor and sensory function was tested by grip strength, incline latency, righting reflex, visual placing, light–dark compartment, rotarod, and open field. These tests confirmed that a ketogenic diet helped improve behavioral abnormalities, specifically a reduction of anxiety when exploring a new environment. This ketogenic diet also helped prevent the onset as well as decreased severity of symptoms associated with this syndrome (Mantis et al. 2009).

The Ketogenic Diet in Depression and Anxiety
Depression is characterized by low mood, loss of interest, and decreased energy. The severity of symptoms is ranked on a spectrum from mild to severe (Sjoberg et al. 2017). Anxiety is described in the Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM-5) as the anticipation of a future threat and is differentiated from a real threat or fear (Crocq 2015). While some believe that pharmacologic methods are adequate to correct these conditions, research shows significant improvements from nonpharmacologic methods, including a ketogenic diet. New studies, including continued on next page
Kraft and Westman (2009) examined a 70-year-old female who was diagnosed with schizophrenia at age 17 based on the presence of auditory and visual hallucinations, paranoia, and disorganized speech. She had been hospitalized multiple times with an increase in psychotic behavior despite heavy doses of antipsychotic drugs. Despite trying multiple combinations of different drugs and dosages, her schizophrenic symptoms persisted, prompting her psychiatrist to suggest nutritional intervention. After being on the ketogenic diet for eight days, she reported an absence of hallucinations and a calmer mood with no disturbances to her medication regimen. After adhering to the ketogenic diet for 12 months, the patient was still experiencing an absence of hallucinations (Kraft and Westman 2009).

Palmer (2017) describes yet another case report where a 33-year-old male had a past medical history of attention deficit hyperactivity disorder, a major depressive disorder, and schizoaffective disorder. He was put on a ketogenic diet to study the effect on the persistent positive and negative symptoms of schizophrenia. This patient had tried multiple medications such as methylphenidate, amphetamine salts, and dextroamphetamine, but symptoms persisted. The patient’s score on the Positive and Negative Symptom Scale (PANSS), which assigns a point value to the schizophrenic symptoms one experiences, was a 98 suggesting “markedly ill”. Within three weeks of a ketogenic diet, the patient reported a decrease in auditory hallucinations, improvement in mood and energy, and a better ability to concentrate. His PANSS score at this time was 49, which is considered “much improved”. A second patient described in this study, a 31-year-old female with a past medical history of major depression and schizoaffective disorder, was also put on a ketogenic diet after seeing no relief with multiple pharmacologic measures and 23 electroconvulsive therapy sessions. Before starting a ketogenic diet, her PANSS score was 107. After four weeks on a ketogenic diet, the patient reported that her delusions were no longer present, and her mood was much better. Her PANSS at this time was 70. After this trial, the patient reported that she stopped the diet and severe paranoia and persecutory delusions followed. Her medication dosage was increased, and she also resumed her ketogenic diet. However, symptoms persisted until she started fasting in order to increase ketosis. After a third day of fasting, her symptoms had resolved (Palmer 2017).

**Conclusion**

While more detailed information is still required on the significance of a ketogenic diet on psychiatric disorders, the studies discussed here encourage nutritional interventions concerning these diseases. If a ketogenic diet is found to be a feasible method of treatment for some psychiatric diseases, psychiatric patients suffering from these diseases may be able to decrease their pharmacologic treatments, which would save money and perhaps reduce the side effects associated with medication use.

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The study of Sussman et al. (2014), suggest that benefits from ketogenic diets can be experienced indirectly as positive effects are passed from the mother to offspring in a mouse model. In this study, dams were started on a ketogenic diet during the mating phase and continued throughout gestation. The pups were placed on a standard diet once weaned from their mother. Magnetic Resonance Imaging (MRI) showed reduced relative volume in the hippocampus, hypothalamus, corpus callosum, striatum, motor cortex, and auditory cortex, and increased relative volume in the cortex and cerebellum. These changes were thought to be related to neurogenesis-induced qualities of ketogenic diets. These brain differences help explain the results discovered during neurobehavioral tests which were performed between eight and 12 weeks of age. Through Open field and Forced Swim Tests, it was found that the prenatal ketogenic diet rodents were less susceptible to anxiety and depression and also experienced increased activity level (Sussman et al. 2014).

Kashiwaya et al. (2013) used a mouse model of Alzheimer’s disease to evaluate the effects of a ketogenic diet on anxiety-like symptoms. The diet was applied to a group of 15 male mice. The ketogenic diet exhibited an anxiolytic effect during Open Field Testing as evidenced by increased ambulation and exploratory behavior compared to the control group (Kashiwaya et al. 2013). Murphy et al. (2004), using a sample of 40 Wistar rats, tested a ketogenic diet in reference to depressive symptoms, or “behavioral despair”. The Porsolt test, a behavioral test, observes rat movement in water and is accepted as a reliable test for depressive symptoms in animals. This study found that during the Porsolt test, rats on a ketogenic diet spent less time immobile than those on a standard diet, meaning they were less likely to exhibit behavioral despair or depression. The results of the rats on a ketogenic diet were similar to rats that had previously been tested while on antidepressants (Murphy et al. 2004).

**The Ketogenic Diet in Schizophrenia**

Schizophrenia is a psychiatric disease that typically arises in adolescence and early adulthood. Diagnosis is associated with structural changes in the brain and abnormal dopamine transmission resulting in hallucinations and delusions (Os and Kapur 2009). A significant problem associated with schizophrenia is referred to as a P50 auditory gate, which occurs when hippocampal interneurons lack the ability to inhibit the response to auditory stimuli, leading to sensory flooding. Research, including the study performed in an analog mouse model of schizophrenia by Tregellas et al. (2015) provides hope that treatments that reduce hippocampal activity can initiate a therapeutic effect in those with schizophrenia. While on a ketogenic diet, a higher level of sensory inhibition was achieved, suggesting that this diet may have a beneficial therapeutic effect on those diagnosed with schizophrenia (Tregellas et al. 2015).

Kraft and Westman (2009) examined a 70-year-old female...
A direct connection between the characteristic symptoms of the psychiatric diseases discussed here and the underlying mechanism of cellular dysfunction offers an excellent opportunity to engage students in a discussion of cell physiology and mitochondrial dysfunction.

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**Literature Cited**
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The Human Microbiome: Composition and Change Reflecting Health and Disease

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Abstract
In humans, microbial organisms known as the microbiota, normally colonize airway passages, skin, the oral cavity, the gastrointestinal tract, and the vagina. There is a growing body of evidence linking the gut microbiota with the overall health of its host. Normally, the microbes that make up the microbiota coexist with a human or animal host without any noticeable difficulty. However, if the symbiotic balance is altered as a result of illness, stress, dietary changes, antibiotic treatment, or other disturbances, the result may be a disruption of normal interactions known as dysbiosis. As a result, the body may become more susceptible to disease. This article examines the most common means by which the microbiota are identified, the process by which the microbiome is acquired, fecal microbial transplantation, and the association of the gut microbiome with specific illnesses such as diabetes and autism. The utility of these applications in developing a teaching module that incorporates the microbiome into courses in physiology or pathophysiology is also reviewed. https://doi.org/10.21692/haps.2019.020

Key words: microbiota, microbiome, HPA axis, dysbiosis, FMT, 16s rRNA gene

Introduction
Healthy adults harbor microbiomes in five major body regions; microbial populations typically inhabit the airway passages, the skin, the oral cavity, the gastrointestinal tract, and the vagina (Proctor 2011). The largest microbiota population is found in the gut where every individual harbors 10 to 100 trillion non-human microbial cells (Figure 1). The study of these cells is relatively new and the terminology applied to them is still evolving. Collectively, the genes of these cells are known as the microbiome and the taxa of human-associated microbes, which include bacteria, bacteriophages, archaea, fungi, eukaryotic viruses and protozoa, are known as the microbiota (Jandhyala et al. 2015; Proctor 2011; Shreiner et al. 2015; Ursell et al. 2012). The vast majority of organisms making up the gut microbiota of adult humans, approximately 1,000 species of bacteria, represent just two bacteria taxa, Bacteroidetes (Gram-negative, non-motile forms) and Firmicutes, most of which are Gram-positive (Jandhyala et al. 2015; Shreiner et al. 2015). The term metagenomics, which was originally used to describe the total DNA present in all of the gut microbes, is increasingly used to refer to specific marker genes, such as the 16s rRNA gene, which is used to study the phylogeny and taxonomy of bacteria. With more than 8,000 species of bacteria named, an increase of over 450% in the number of described taxa since 1980, the field of bacterial genetics, particularly the genetics of the organisms related to the human microbiome, is exploding (Janda and Abbott 2007).

Figure 1. The microbiome includes 10 to 100 trillion non-human microbial cells including bacteria, bacteriophages, archaea, fungi, eukaryotic viruses and protozoa. Depositphotos, 2019. Open access.
Antonie van Leeuwenhoek is credited with carrying out the first exploration of the human microbiome in the early 1680s when he compared his own fecal and oral bacteria. He observed and recorded the differences among the microbes in these two habitats and also noted the differences that he observed in samples taken from these sites in healthy and diseased individuals (Ursell et al. 2012). In keeping with the progression of scientific research since Leeuwenhoek’s day, the primary focus of today’s research is not merely to observe the obvious differences among site-specific microbial populations, but rather to use advanced molecular techniques to help explain why these population differences exist and to illuminate the physiological significance of the observed populations (Ursell et al. 2012).

The composition of the microbiota of the mammalian gut is unique to each individual and very different from free-living microbial communities. For example, bacteria associated with extreme external environments, such as thermoacidophils, are similar to communities in other environments, while mammalian gut bacteria appear to represent an extreme case of microbial existence confined to a specific internal environment that is warm, food rich, and relatively stable (Ursell et al. 2012).

As catalogued by the European Metagenomics of the Human Intestinal Tract (Meta-HIT) consortium in 2010, there may be as many as 3.3 million non-redundant genes in the microbiome of the human gut, compared with approximately 22,000 genes in the entire human genome (Shreiner et al. 2015; Ursell et al. 2012). The genetic diversity of the microbiome in any individual is immense. While individual humans are typically approximately 99.9% genetically identical to each other, each individual’s gut microbiome can be as much as 80-90% different from all others. This suggests that, when technology and information become available, targeting the variations of the microbiome for personalized medical treatment may ultimately prove to be more efficacious than targeting the relatively consistent DNA of the human host genome (Ursell et al. 2012).

This article examines the use of the 16s rRNA gene in identifying the microbiota, the process of acquiring a microbiome, fecal microbial transplantation, and the association of the gut microbiome with diabetes, autism, stress, and the hypothalamic-pituitary-adrenal axis. The utility of these applications in developing a teaching module that incorporates the microbiome into courses in physiology or pathophysiology is also reviewed.

The use of 16s rRNA gene in identifying the microbiota.
The various species that comprise the human microbiota are most commonly identified by sequence analysis of the 16s rRNA gene (Gill et al. 2006; Nasidze et al. 2009; Lagier et al. 2012; Earl et al. 2018; Park et al. 2019). This highly conserved gene is ~1550 bp long and is transcribed to produce 16s RNA, a critical component of the small ribosomal subunit found in all prokaryotes. Sequence analysis of the 16s rRNA gene has been useful in assessing phylogenetic relationships (Woese 1987; Clarridge et al. 2004). The gene includes highly conserved, variable, and hypervariable regions, which mutate at different rates and are quite diverse among different species of prokaryotes (Ludwig and Schleifer 1994; Van de Peer et al. 1996; Chakraborty et al. 2007). As a result, sequence analysis of the 16s rRNA gene can be used to identify the individual organisms comprising the microbiota using the readily available 16s ribosomal databases that include SILVA (Quast et al. 2013), RDP (Wang et al. 2007), and Greengenes (McDonald et al. 2012). Most studies using this approach are able to classify organisms from phylum through genus, but only sometimes to the level of species (Gill et al. 2006; Chu et al. 2010; Yarra et al. 2014).

There is no generally agreed upon species concept for Bacteria and Archaea (Cohan 2001; Dykhuisen 2005; Staley 2006; Achtman and Wagner 2008). Eukaryotic species are typically defined as populations of morphologically similar organisms that are able to interbreed and that are reproductively isolated from other such populations (Sokal and Crovello 1970; Mayr 2000). However, this species concept is not easily applied to prokaryotes. Of the estimated millions of different bacteria (Curts et al. 2002), most have not been cultured and are only known by DNA sequence (Venter et al. 2004; Giovannoni and Stinge 2005). As a result, “operational taxonomic units” (OTUs) are commonly used to approximate bacterial species. Typically, OTUs are characterized as those with a minimum of 97% 16s rRNA sequence similarity (Stackebrandt and Goebel 1994; Schloss and Handelsman 2006; Schloss 2010; Koeppel and Wu 2013).

There are a number of different approaches to sequencing 16s RNA. The first methods of DNA sequencing involved chemical cleavage and electrophoresis (Maxam and Gilbert 1977) or primer directed synthesis and chain termination (Sanger et al. 1977). With the development of capillary gel electrophoresis (Swerdlow and Gesteland 1990; Luckey et al. 1990) and automated technologies (Wilson et al. 1988; D’Cunha et al. 1990), Sanger (chain termination) sequencing has become the standard method for sequence analysis and, due to its low error rate and read lengths of 700 bp or higher, remains the gold standard in applications that do not require a high throughput. The advent of next generation sequencing (NGS) or second-generation sequencing resulted in the ability to analyze a much larger number of sequences (millions of reads per sample) at a lower cost, although read lengths tend to be shorter than with Sanger sequencing (Caporaso et al. 2011; Slatko et al. 2011).
2018). Newer third and fourth generation technologies such as nanopore sequencing (Branton et al. 2008; Liu et al. 2016) produce much longer read lengths with high throughputs but also with higher error rates (Schadt et al. 2010; Slatko et al. 2018). These newer technologies allow for full length sequencing of 16s rRNA, so this approach, coupled with NGS, is expected to be useful in the accurate identification of microbial diversity (Bashir et al. 2012; Shin et al. 2016).

**How does an individual acquire a microbiome?**

Human babies in utero do not possess a microbiome. Newborns are colonized by microbiota during the birth process. Within twenty minutes after delivery, the microbiota of infants delivered vaginally is markedly similar to the microbiota of the mother’s vagina. Infants who are delivered by Cesarean section have microbiota that are typically associated with the skin (Barko et al. 2018; Fuentes et al. 2016; Ursell et al. 2010). Babies continue to acquire microbiota during the first few years of life and by the time they are approximately a year old, the microbiome of the digestive tract begins to resemble that of the adult gut microbiome (Barko et al. 2018; Fuentes et al. 2016; Ursell et al. 2010).

As the baby grows, there is a steady increase in the phylogenetic diversity of the gut microbiome. Changes in the composition of the microbiota of babies have been observed to occur in tandem with dietary changes such as the introduction of breast milk, the addition of rice-based cereal, the introduction of formula, and eventually, the introduction of table food (Jandhyala et al. 2015; Ursell et al. 2012). For example, when babies transition to an adult diet, genes associated with vitamin synthesis and the digestion of polysaccharides typically make an appearance (Ursell et al. 2012).

**Changes in the composition of the gut microbiome**

The gut microbiome is not a static entity. It changes and adapts over the course of a person’s lifetime. Changes in the microbiome are associated with antibiotic treatments and with diseases such as inflammatory bowel disease, obesity, asthma, metabolic syndrome, cardiovascular disease, immune related conditions and even autism spectrum disorder (Barko et al. 2018; Proctor 2011; Ursell et al. 2012).

Antibiotics have a significant impact on the composition of the gut microbiome. For example, treatment with ciprofloxacin, a broad-spectrum antibiotic that functions as an inhibitor of the bacterial enzyme topoisomerase, results in a decrease in the number of taxa and the diversity of the gut microbiota within three to four days (Shreiner et al. 2015). Typically, the gut microbiota begin to resemble the pre-treatment state approximately a week after antibiotic treatment is stopped but there are significant individual differences in how long the process of returning to normal takes and sometimes, certain taxa do not return to the internal community at all. In extreme cases, it can take up to four years following treatment with antibiotics for some microbiota species to become re-established (Jandhyala et al. 2015; Shreiner et al. 2015; Ursell et al. 2012). Due to ethical issues associated with administering antibiotics to healthy people the mechanisms by which the gut microbiome is replenished after a major disruption and the substantial differences in the process from person to person have not yet been fully described (Shreiner et al. 2015; Ursell et al. 2012).

**Fecal microbial transplantation and the treatment of Clostridium difficile infection.**

*Clostridium difficile* infection (CDI) is a human disease that develops as a direct result of changes in the gut microbiome (Shreiner et al. 2015; Ursell et al. 2012). *Clostridium difficile* is a Gram-positive, spore-forming, anaerobic bacterium of the taxa *Firmicutes* that may carry antibiotic resistance and is known to produce toxins that are associated with severe diarrhea and colitis (Fuentes and de Vos 2016). CDI is the most common cause of nosocomial antibiotic–associated diarrhea with approximately 3,000,000 cases reported each year (Cole and Stahl 2015). The infection can sometimes be cured with oral and intravenous antibiotics but an increased incidence of the disease and the appearance of more virulent strains have led to more cases of persistent, recurrent, or relapsing CDI, which can lead to years of escalating health care problems and death. CDI spores may persist for long periods on contaminated hospital surfaces and may be carried by health-care workers. In fact, these spores are so hardy that alcohol-based antiseptics are not enough to effectively kill them and hands must be washed with chlorhexidine soap to decrease the risk of spreading the infection (Cole and Stahl 2015). Risk factors include being older than 65, being treated with antibiotics, having a severe illness, and being hospitalized (Cole and Stahl 2015). CDI is characterized by a disturbed, low-density community of microbiota that is antibiotic induced (Fuentes and de Vos 2016). Fecal microbial transplantation (FMT) is currently the most successful treatment option for this disease.

Fecal microbiota transplantation is a straightforward procedure in which healthy microbiota from a donor individual are transferred directly into the gastrointestinal (GI) tract of a disturbed, dysbiotic microbial community of a recipient in order to re-establish the normal gut microbiota (Barnes and Park 2017; Fuentes and de Vos 2016; Shreiner at al. 2015; Ursell 2012). The microbiota can be transferred in pill form, via gastronomy or nasogastric tube into the stomach, post-pylorically (distal placement of a gastronomy or nasogastric tube into the duodenum or jejunum), colonoscopically, or via enema (Barnes and Park 2017). The data do not yet support a superior means of microbiota introduction.

For successful FMT, the patient’s GI tract undergoes bowel lavage or treatment with laxatives prior to the procedure.

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Stool samples are screened for parasites and a variety of pathogenic bacteria and blood samples are screened for transmittable diseases such as hepatitis and HIV. Donor questionnaires, similar to those used for blood bank donors, are used to screen for non-GI diseases that might prove to be problematic (Barnes and Park 2017; Fuentes and de Vos 2016; Shreiner at al. 2015; Ursell 2012).

FMT therapy is known variously as fecal bacteriotherapy, fecal transfusion, duodenal infusion, probiotic infusion and even, since the work of Petrof et al. (2013), as “rePOOPulating” (Fuentes and de Vos 2016; Petrof et al. 2013). The majority of published data describing FMT come from studies of the treatment of CDI but the process is being investigated to treat ulcerative colitis, acute graft-versus-host disease, and diseases of the bowel such as inflammatory bowel disease and irritable bowel syndrome (Barnes and Park 2017). FMT is also being considered as treatment for non-bowel diseases such as multiple sclerosis, metabolic syndrome, and diabetes (Barnes and Park 2017; Proctor 2011).

The first recorded FMT procedure is credited to the work of Benjamin Eisman and his colleagues in 1958, although the practice may date back 1700 years to the Djong-ji dynasty (Fuentes and de Vos 2016; Khoruts 2017). Eisman et al. (1958) reported a case study of four patients with pseudomembranous enterocolitis, a condition caused by *Clostridium difficile*, who were cured with “fecal enemas”. He noted that in the early days of oral antibiotics, physicians frequently encountered cases of severe diarrhea due to the over-use of antibiotics and lack of standard dosing protocols (Khoruts 2017). Pseudomembranous colitis was a terrible disease in the 1950’s with a mortality rate approaching 75%. The treatment of choice was surgical removal of the colon, which is still done in cases of antibiotic-refractory severe-complicated CDI infections and, even today, this surgery carries a mortality rate of approximately 50% (Khoruts 2017). Using FMT, the cure rate for CDI may be as high as 91% with the resulting creation of a stable bowel homeostasis that prevents further infections (Fuentes and de Vos 2016).

One of the goals of current research in this area is to identify specific communities of microbiota that can be used successfully and predictably for treating specific diseases. Another goal is to develop synthetic microbial communities that can be reproduced without human donors and stocked for future off-the-shelf use (Fuentes and de Vos 2016; Shreiner et al. 2015; Ursell 2012). Ready-to-use solutions of microbiota are currently being prepared and tested for commercial use in the treatment of recurrent CDI. If they become widely available, synthetic microbiota solutions for treatment of a range of specific diseases would overcome the problem of constantly screening for ideal donors (Barnes and Park 2017). If at some point in the future FMT becomes an easier, less invasive, more esthetically acceptable process, perhaps with the use of encapsulated cryopreserved microbiota, it may one day help to reduce the use of antibiotics such as vancomycin in the treatment of multidrug-resistant organisms that reside in the bowel (Crum-Clanflone et al. 2015).

The Food and Drug Administration (FDA) classified human feces for medical use as a drug in 2013, in an attempt to regulate and ensure the safety of fecal transplantation. The action was prompted in part by the appearance of do-it-yourself online treatment videos appearing on the Internet (Fuentes and de Vos 2016).

In spite of the enormous success of FMT treatment, the process may have side effects, including abdominal pain, bloating, nausea, and vomiting; most of which are mild. More severe side effects may include post-transplant sepsis and intestinal perforation (Fuentes and de Vos 2016).

Further studies of FMT are needed to evaluate the potential risk with respect to the transmission of autoimmune diseases, metabolic diseases, and cancer. The primary concern with respect to FMT is the possible transmission of resistant bacteria, unknown viruses, or other as yet unrecognized infectious agents, from donor to recipient (Fuentes and de Vos 2016). The FDA issued a warning on June 13, 2019 following the death of a FMT recipient from an invasive extended-spectrum beta-lactamase (ESBL) infection that was caused by resistant *E. coli* (Yancey-Bragg 2019). The patient who died was known to be immune compromised and the donor stool had not been tested for drug-resistant bacteria prior to the transplant. After the patient died, the stored donor stool sample was tested and found to contain the resistant *E. coli*. The FDA now requires that all donor stool samples be tested for drug resistant bacteria in addition to standard pre-transplant testing (Yancey-Bragg 2019).

**Diabetes and the microbiota of the gut**

Diabetes has evolved into a potentially deadly public health epidemic of pandemic proportions, with a disease prevalence approaching ten percent globally (Aw and Kukuda 2018). Type 1 diabetes (T1D) is an autoimmune disease that occurs when T cells of the immune system attack pancreatic islet β-cells rendering the pancreas incapable of producing adequate amounts of insulin. T1D accounts for 10% of all cases of diabetes (Tai et al. 2015). Type 2 diabetes (T2D) is a complex metabolic disorder that includes insulin resistance, in which the body cannot effectively utilize the insulin it produces. Insulin resistance is associated with obesity. Genetic factors are known to be critical in the pathogenesis of diabetes and there is growing evidence that the gut microbiota may also be important in influencing the development of T1D and T2D (Tai et al. 2015).

Several studies have suggested that short chain fatty acids (SCFAs) play a role in the pathogenesis of T2D (Aw and Fukuda 2018). These studies report that the number of bacteria that
produce SCFAs is lower in people with T2D diabetes. This is problematic because SCFAs, adhering to G-protein coupled receptors, which play a role in regulation of lipid and glucose metabolism, are known to have wide ranging effects in the body. For example, SCFAs promote secretion of glucagon-like peptide-1, which impedes secretion of glucagon, slow down gluconeogenesis in the liver, increase insulin sensitivity, and increase central satiety, which may result in weight loss (Aw and Fukuda 2018). SCFAs can also disrupt the low-grade inflammatory response caused by bacteria moving from the intestines into the surrounding adipose tissue and the blood (Aw and Fukuda 2018).

Lifestyle changes may have played a role in the increased incidence of diabetes in the United States over the last 50 years (Aw and Fukuda 2018; Ursell et al. 2012). During this period antibiotic consumption has increased and the Western diet has changed. Antibiotic consumption may result in a decrease in the overall diversity of the gut microbiota, which has been implicated in reduced immune function (Aw and Fukuda 2018; Ursell et al. 2012). A reduction in the diversity of the gut microbiota has also been observed in T2D and recent studies have shown that people with T2D are less likely to have the gut bacteria that digest plant polysaccharides into SCFAs. The presence of these bacteria in healthy patients facilitates the production of SCFAs (Aw and Fukuda 2018; Ursell et al. 2012). A change in eating habits, primarily an increased dependence on highly processed food, has resulted in an increase in the consumption of carbohydrates and fats for many people, accompanied by a decrease in the consumption of dietary fiber to less than half of the recommended fiber intake of 30 g. daily (Aw and Fukuda 2018). Normally, indigestible fiber is fermented in the digestive system by organisms present in the gut microbiota, producing SCFAs that exert an anti-inflammatory effect by producing immunoglobulin A and immunosuppressive cytokines (Aw and Fukuda 2018). A diet reduced in fiber is associated with a decrease in the number of SCFAs and the microbes capable of synthesizing them. A diet high in fiber may help to maintain the bacteria needed to produce SCFAs. Loss of diversity of the microbiota and a decrease in the number of SCFAs, result in dysbiosis that is implicated in an increase in the presence of inflammatory diseases, including diabetes (Aw and Fukuda 2018; Ursell et al. 2012).

A recent study on human males with T2D revealed that they had fewer bacteria of the taxa *Firmicutes* when compared to a non-diabetic population and more *Betaproteobacteria*, some of which may be pathogenic (Aw and Fukuda 2018). This study also identified positive correlations between plasma glucose levels and the *Bacteroidetes* to *Firmicutes* ratio (which normally changes over a person’s life time) as well as the ratio of the *Bacteroides*–*Prevotella* group (associated with a plant-rich diet and a high protein diet respectively) to the *Clostridium cocoides*–*Eubacterium rectale* group, which produces butyrate (Aw and Fukuda 2018; Prados 2016). Butyrate is a SCFA found in the colon. It has several known functions; it serves as the preferred energy source for epithelial cells of the colon, enhances the barrier function of the gut, and enhances the immune and anti-inflammatory properties of the gut (Riviere et al. 2016). The reduction of butyrate-producing bacteria in the gut microbiota is correlated with impaired insulin sensitivity and obesity and it may contribute to the disease pathology in people with T2D diabetes (Aw and Fukuda 2018). The Aw and Fukuda (2018) study suggests that Gram-negative *Bacteroidetes* and *Proteobacteria* might be implicated in the pathogenesis of T2D through an inflammatory response induced by endotoxins (Aw and Fukuda 2018). If further research supports this conclusion, the gut microbiome might become a reliable biomarker for predicting the onset of T2D in glucose-intolerant people. It has also been noted that treating people with metabolic syndrome with vancomycin reduces the number of Gram-positive butyrate-producing bacteria.

In studies of children who are at high risk for developing T1D due to the presence of islet autoantibodies, the gut microbiota has been shown to be consistently less diverse and less dynamic than that of healthy controls (Tai et al. 2015). It is characterized by low numbers of lactate- and butyrate-producing bacterial species and very low numbers of the two most prominent *Bifidobacterium* species, *Bifidobacterium adolescentis* and *Bifidobacterium pseudocatenulatum*, which are common inhabitants of the gut microbiota that are often used as probiotics (Tai et al. 2015). The disturbed microbiota in children at risk for developing T1D also displayed an increased presence of bacteria of the taxa *Bacteroides*, which normally function in the breakdown of complex organic molecules (Tai et al. 2015). These studies suggest that changes in the gut microbiota are associated with T1D and that perhaps targeted FMT to re-establish a more normal microbiome might one day be used to delay and/or prevent the onset of diabetes (Tai et al. 2015).

Further research is needed to determine the mechanisms by which the altered microbiota of the gut develop in people with diabetes and the manner in which the immune system might interact with the microbiota with respect to this disease (Tai et al. 2015). It has not yet been determined whether the gut microbiota initiate the development of diabetes, enhance the disease once it is present, or are a result of the disease (Tai et al. 2015).

The microbiome in stress and mental health

Stress is known to affect the physiology of the GI tract in several ways including: changes in motility and secretions, increased permeability, decreased ability to maintain the mucosal lining, and changes in the gut microbiota (Konturek et al. 2011). There is a growing body of empirical evidence to support a possible connection between the gut microbiome, stress, and cognitive function, primarily in the expression of depression and anxiety (Liu 2016). Several studies have found that psychosocial stressors may change the composition of the gut microbiota, resulting in the increased presence of cytokines, which have

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been associated with activation of the vagus nerve and the risk for psychiatric disorders such as depression (Dowlati et al. 2010; Liu 2016; Mills et al. 2013).

Support for a possible connection between the gut microbiota and stress-related psychiatric illness comes partly from demonstrating the anxiety-inducing effects of certain bacteria and the anti-depressive or anti-anxiety effects of probiotics in rodents (Liu 2016). For example, increases in anxiety-related behavior has been observed in rodents exposed to Citrobacter rodentium (the rodent equivalent of E. coli), Trichuris muris (a nematode parasite of mice), and Campylobacter jejuni, the most commonly reported cause of bacterial food poisoning in the US (Altekruse et al. 1999; Lyte 1998; Liu 2016), while Lactobacillus spp. and Bifidobacterium spp. (probiotics) have been demonstrated to reduce anxiety and depression-like behavior in rodent studies (Arseneault-Bréard et al. 2012; Lui 2016).

Very few human studies have been done in this area; however, two recent observational studies have examined the gut microbiota with respect to human psychopathology. In one study, adult patients with clinical depression were found to have higher levels of Enterobacteriaceae and Alistipes and lower levels of Faecalibacterium relative to healthy controls. Enterobacteriaceae is a large taxa of bacteria that includes E.coli, Alistipes species are associated with the serotonin precursor tryptophan, and Faecalibacterium is known to have anti-inflammatory properties (Jiang et al. 2015). In the second study it was found that depressed adults had higher concentrations of Bacteroidales, the most abundant beneficial gram-negative organism in the human gut microbiota, and lower concentrations of Lachnospiraceae, an anaerobic bacteria that is believed to be protective against colon cancer by producing butyric acid (Coyne and Comstock 2008; Meehan et al. 2014; Naseribafrouei et al. 2014; Lui 2016). The results of these studies are consistent with the possibility that there are changes in the microbiota that accompany psychological disorders. More research with psychiatric populations will need to be done in order to determine the relevance of these differences (Lui 2016).

Several studies have examined the use of probiotics or prebiotics in humans. In a representative study, participants given a combination of the probiotics Lactobacillus and Bifidobacterium species, demonstrated lower scores on measures of psychological distress, hostility, anxiety, and depression compared to a group given a placebo (Messauodi et al. 2011). These studies are limited by the fact that the investigations were confined to psychiatrically healthy participants (Liu 2016). Further research is needed to directly evaluate the relation between the gut microbiota and clinically significant psychopathology.

While research on the microbiome continues, adopting a program to manage chronic stress may be one way to positively impact the gut microbiota. Mindfulness and other meditation practices are now considered standard methods to build resistance to the impact of stress on the body (Bergland 2016; Cooper et al. 2018). Additionally, exercise is a well-known approach to lowering blood pressure and cortisol levels and increasing overall feelings of well-being and there is evidence to support a relationship between the diversity of the gut microbiota and exercise (Fard 2014; Georgia State 2018). While more studies are needed to confirm the exact relationship between stress management and the microbiome, the impact of stress on the body and the potential benefits of mindfulness and exercise for overall health are well documented and undeniable (Cooper et al. 2018).

**Autism spectrum disorder and the microbiome**

Autism Spectrum Disorder (ASD) is a range of disorders affecting one in 59 children at age eight. It is an intensely researched disorder but the etiology remains unclear and there is, as yet, no approved treatment for the core symptoms (Baio et al. 2014). While social and repetitive behavioral abnormalities are among the most recognizable symptoms of ASD, patients also demonstrate elevated levels of inflammation and gastrointestinal abnormalities. Mounting evidence indicates that the microbiome may play a role in the development and severity of symptoms in ASD, and possibly in the causality of ASD in young children (McElhanon et al. 2014).

Several studies have demonstrated differences in the gut microbiome of ASD as compared to neurotypical children, including reductions in fermenting bacteria such as Prevotella and increased levels of the propionic acid-producing bacteria Clostridia (Song et al. 2004, Finegold et al. 2010, Kang et al. 2013). Propionic acid has been shown to lower the fatty acid content of the liver and blood plasma, reduce food intake, and improve insulin sensitivity. Consequently, an increase in propionic acid production may be associated with the prevention of obesity and T2D (Al-Lahham et al. 2010). Propionic acid may also increase the overall threshold for inflammatory responses and it may have a major role to play in the link between nutrition, the gut microbiota, and human physiology (Al-Lahham et al. 2010).

The differential microbiomes associated with ASD may impact the development and function of the nervous system through enrichment or depletion of specific neuroactive microbial metabolites. Specifically, the SCFAs produced by some of these microbes impact the maturation and behavior of microglia (Enry et al. 2015). Microgla, which have several functions in the brain including phagocytosis of pathogens and the removal of damaged cells, are known to be overly activated in patients with ASD (Pardo et al. 2005). They may be associated with the neuroinflammation that is present in ASD as well as in inappropriate developmental pruning, the process of eliminating extra synapses that is widely thought to be

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associated with learning and efficient functioning of the brain (Paolicelli et al. 2011).

Several promising studies have suggested that current research in the role of the microbiome in ASD may result in therapies for ASD patients. Fecal transplants in ASD patients have resulted in decreases in GI, social, and behavioral symptoms (Kang, Adams et al. 2017) that have continued for as long as two years after the initial transfer of microbiota (Kang et al. 2019). More specific treatments may include the transfer of a single species of microbe, such as *Bacteroides fragilis*, a human commensal organism that has been shown to improve behavioral abnormalities in a rodent model of ASD (Hsiao et al. 2013).

Not all proposed treatments for ASD rely on microorganisms since they can be unstable due to host-organism interactions with the patient. However, treatment of animal models of ASD with specific microbiome metabolites such as 5-aminovaleric acid (5AV) and the amino acid taurine, both of which affect gamma-aminobutyric acid (GABA) receptors in the brain and are known to be reduced in ASD, has been shown to reduce repetitive ASD-like behaviors and increase social behaviors in mice (Sharon et al. 2019). More research is needed determine whether clinically significant metabolites such as 5AV and taurine exist in human patients with ASD and if targeted metabolites produced by microbiota might become effective treatment therapies for disorders that are seemingly centered in the brain.

The effect of gut microbiota on the hypothalamic-pituitary-adrenal (HPA) axis during stress

The human microbiome has been linked to the regulation of several physiological processes including those that are altered under stressful conditions in order to maintain homeostasis (Frodl and O’Keane 2013; Luczynski et al. 2016). The hypothalamic-pituitary-adrenal (HPA) axis is a neuroendocrine bidirectional communication pathway between the gut and the central nervous system that produces cortisol. It is an important component of the stress management homeostatic system (Cussotto et al. 2018; Frodl and O’Keane 2013). Cortisol is a glucocorticoid that alters several tissues in order to mobilize or store energy under stress conditions (Frodl and O’Keane 2013). Glucocorticoids bind to glucocorticoid receptors (GRs) in the hippocampus and mineralocorticoid receptors (MRs) dispersed throughout the brain; both receptors function as transcription factors (Frodl and O’Keane, 2013). Cortisol has a higher affinity for the MRs than GRs, which allows MRs to help maintain the low cortisol levels in the blood under normal conditions (Stevens and Wand 2012). Under stress conditions, higher concentrations of cortisol cause the cortisol to bind to GRs with lower affinity, which ultimately causes GRs to terminate the stress response as a result of a negative feedback reaction that is described below (Stevens and Wand 2012). The modulation of glucocorticoids plays an important role in maintaining normal function of the HPA axis (Frodl and O’Keane 2013).

Under stressful conditions, the HPA axis causes the release of corticotropin-releasing hormone (CRH) from the paraventricular nucleus of the hypothalamus (Frodl and O’Keane 2013). CRH is then carried to the anterior pituitary gland where it binds to its receptors and causes the secretion of adrenocorticotropic hormone (ACTH) into peripheral circulation (Frodl and O’Keane 2013). ACTH causes the release of cortisol (in humans) and corticosterone (in mice) from the adrenal gland, which leads to negative feedback of ACTH and CRH secretion and restoration of homeostasis (Figure 2) (Frodl and O’Keane 2013). The negative feedback of these hormones helps prevent prolonged activity of the HPA axis (Stevens and Wand 2012). Imbalances in the HPA axis have been associated with mood and anxiety disorders as well as digestion, immunity, emotions, sexuality, and energy expenditure (Cussotto et al. 2018).

![Figure 2. The HPA axis is responsible for the neuroendocrine adaptation component of the stress response. Depositphotos.com](image)
In investigating the link between gut microbiota and the HPA axis during stress conditions, germ-free (GF) and specific-pathogen-free (SPF) mice are commonly used (Luczynski et al. 2016). GF mice are raised in an environment without exposure to microorganisms, while SPF mice have normal microbiota but are guaranteed to be free of certain pathogens (Luczynski et al. 2016). Sudo et al. (2004) undertook the first major study that linked gut microbiota to the stress response by comparing HPA axis activity of GF and SPF mice using an acute restraint stress protocol. The results of the study showed that GF mice under stress displayed higher levels of plasma ACTH and corticosterone compared to SPF mice (Sudo et al. 2004). The study concluded that the presence of gut microbiota influences normal regulation of the HPA axis (Sudo et al. 2004).

Clarke et al. (2013) observed the effect of gut microbiota on the serotoninergic system, which is known to be associated with stress and anxiety. The study compared GF and SPF mice under environmental stress, which can include conditions such as isolation, noise, and low temperature (Clarke et al. 2013). The results of the study showed that GF mice had increased levels of 5-hydroxytryptamine (serotonin) and 5-hydroxyindoleacetic acid, the primary metabolite of serotonin. Serotonin is associated with feelings of well-being and happiness as well as reward, memory, learning, and other physiological processes and 5-hydroxyindoleacetic acid is the marker detected in urine that is used to determine the amount of serotonin present in the body. In addition, tryptophan (a precursor of serotonin) was also higher in GF mice (Clarke et al. 2013).

Diaz Heijtz et al. (2011) observed motor activity and anxiety-like behavior in GF and SPF mice using three types of stress tests: open-box, light-dark box, and elevated plus maze. The open box stress test is based on the preference rodents display for hidden, enclosed spaces. The light-dark box stress test is based on the aversion rodents display to brightly illuminated spaces and the elevated maze test is based on their aversion to open spaces. Rodents normally exhibit locomotion behavior in response to novel environments, which means they will move freely in a new environment seeking the place of least stress. During a typical light-dark box experiment, for example, the amount of time an animal spends transitioning from a light to a dark environment might be recorded. The results of the study showed that GF mice displayed decreased anxiety-like behavior and increased motor activity compared to SPF mice. The GF mice also displayed altered expression of genes related to anxiety and synaptic plasticity (Diaz Heijtz et al. 2011). Similarly, Neufield et al. (2011) compared the behavior of stressed GF and SPF mice using an elevated plus maze test. The study found that GF mice exhibited less anxiety-like behavior than SPF mice and increased levels of plasma corticosterone (Neufield et al. 2011). The results of these studies suggest that increased anxiety-like behavior in mice may be due to hyperactivity of the HPA axis and modulation of anxiety-related genes (Diaz Heijtz et al. 2011; Neufield et al. 2011). Overall, these studies suggest that the presence of gut microbiota play a key role in normal behavioral response to stress (Diaz Heijtz et al. 2011; Neufield et al. 2011).

Two of the most recent studies, Huo et al. (2017) and Vodička et al. (2018), suggest that the absence of gut microbiota can alter the activity of the HPA axis, highlighting the importance of the gut-brain axis. When GF mice are compared to SPF mice, these studies revealed clear differences in how certain functions related to the HPA axis are either increased or decreased under stress conditions (Huo et al. 2017; Vodička et al. 2018). From the findings of the Huo and Vodička studies, it can be concluded that gut microbes modulate the HPA axis by impacting anxiety-like behavior, normal levels of HPA axis hormones, and the expression of hormone receptors and genes (Huo et al. 2017; Vodička et al. 2018).

Vodička et al. (2018) observed that GF mice spent less time in total defensive behavior compared to SPF mice and that SPF mice displayed more escape/flight behavior compared to GF mice when faced with a social defeat procedure during which mice are subjected to prolonged social stress by being exposed to large, aggressive mice (Vodička et al. 2018). Escape/flight, which is defined as running or jumping away from resident mice, is an example of anxiety-like behavior. The results of this study suggest that GF mice are less likely to exhibit anxiety-like behavior under stress conditions (Vodička et al. 2018). Huo et al. (2017) found that stressed GF mice traveled a greater distance in an open field test, where they were permitted to move freely, and spent more time in the center of the open field test apparatus compared to stressed SPF mice (Huo et al. 2017). Greater traveling distance and expenditure of time in the central area are examples of anti-anxiety like behavior in mice (Huo et al. 2017). Although a few studies have shown an increase in anxiety-like behavior in GF mice, the majority of studies have found a decrease in this response and indicated it as an impaired behavioral response to stress (Rabot et al. 2016). The contrast in these results may be due to differences in methodology or the effect of genetic background of the mice (Rabot et al. 2016). It is important to note that the behavioral response results in the Vodička et al. (2018) study correlated with hormone changes of the HPA axis (Huo et al. 2017). Stressed GF mice showed less anxiety-like behavior along with over-activity of the HPA axis (Huo et al. 2017). This finding suggests that gut microbiota may regulate anxiety-like behavior through an endocrine response (Huo et al. 2017).

The normal regulation of HPA axis hormone levels is also impacted by gut microbiota (Huo et al. 2017). The regulation of these hormones is important since corticotropin-releasing hormone (CRH) causes secretion of ACTH, which leads to the release of cortisol (Frodl and O’Keane 2013). Huo et al. (2017) found that the absence of gut microbiota caused the upregulation of both CRH and ACTH in stressed GF mice compared to stressed SPF mice (Huo et al. 2017). This finding continued on next page
was similar to the Sudo et al. (2004) study of GF and SPF mice subject to stress. In addition, the study found an increase in cortisol (CORT) and aldosterone (ALD) levels (Huo et al. 2017). There is debate about whether corticosterone levels are changed in GF mice since some studies find an increase in concentration while others observe a normal range (Luczynski et al. 2016). These contrasting results may be due to the type of microbiota examined, the sex of the mice, or differences in the experimental procedure (Luczynski et al. 2016). Overall, the increase in these hormones in the Huo et al. (2017) study may explain the decreased anxiety-like behavior observed in stressed GF mice, whereas stressed SPF mice exhibited normal behavior following a stress protocol (Huo et al. 2017).

Gut microbes regulate the expression of hormone receptors and genes under stress conditions (Huo et al. 2017; Vodička et al. 2018). In the Huo et al. (2017) study, it was found that GF stressed mice had increased levels of CRH receptor Type 1 (Crhr1) mRNA and decreased levels of Nr3c2 mRNA, which encodes the mineralocorticoid receptor that regulates the action of aldosterone (Huo et al. 2017). Huo et al. (2017) also found that GF stressed mice had decreased MR/GR (Huo et al. 2017). Previous studies have suggested that changes in MR/GR expression may indicate HPA axis dysfunction in mood-related disorders (Webster 2002). Overall, the results from the Huo et al. study suggest hormone dysfunction in the hippocampus of GF mice under stress (Huo et al. 2017).

In addition, Vodička et al. (2018) found that stress increased the expression of proopiomelanocortin (POMC), the pituitary precursor of melanocyte stimulating hormone and ACTH, but did not significantly affect corticotropin releasing hormone receptor 1 (Crhr1) (Vodička et al. 2018). Since POMC is a precursor of ACTH, its increased levels matched the increase in ACTH in the Huo et al. (2017) study (Huo et al. 2017; Vodička et al. 2018).

Vodička et al. (2018) also found that the expression of the Fkbp5 gene in the pituitary gland was decreased in SPF mice and upregulated in GR mice. The function of the Fkbp5 gene is to encode a protein that controls negative feedback by decreasing the affinity of GR for corticosterone (Vodička et al. 2018). It is suggested that higher expression of Fkbp5 in GF mice may result in decreased efficiency of negative feedback via GR (Vodička et al. 2018). Therefore, the upregulation of Fkbp5 in GF mice may be one reason that an exaggerated HPA response was observed in these mice (Vodička et al. 2018). This finding correlates with previous studies that have shown an increased expression of Fkbp5 and the GR levels in the cytoplasm during chronic mild stress (Guidotti et al. 2013). The results of the Vodička et al. (2018) study also suggest that the absence of microbiota increased the expression of genes encoding proteins involved in steroidogenesis (Star and Cyp11a1) and biosynthesis of catecholamines (TH and PNMT) in the adrenal gland (Vodička et al. 2018). In contrast, stress only affected genes encoding epinephrine synthesis (Vodička et al. 2018). These findings suggest that the presence of microbiota help regulate normal catecholamine biosynthesis and steroidogenesis, and the absence of these microbes can lead to dysregulation of these processes (Vodička et al. 2018).

Further research is needed to provide much needed knowledge about how the gut microbiota influence and interact with the function of the HPA axis and stress-related disorders (Huo et al. 2017; Vodička et al. 2018).

**Conclusion**

An unprecedented amount of growth in our understanding of the microbiome has come about relatively quickly as a result of the availability of powerful research tools, but there are still many questions that remain unanswered. Emerging technologies such as 16s rRNA sequencing and FMT raise the prospect of a future in which specific treatments for diseases such as diabetes, metabolic syndrome, and mental disorders can be targeted to individual humans. Determining the relationships between the gut microbiota, systemic diseases and diseases of the bowel, stress-related mental illnesses, and physiological processes such as those that function within the HPA axis, is clinically extremely important. Preliminary evidence suggests that the use of probiotics and FMT may be free of some of the side effects and addictive properties that are associated with pharmacological medications; this bodes well for their potential safety and tolerance as a form of treatment. As always, inherent in the world of today is an ever-present need for the development of new treatment options for the future.

The information contained in this article provides a physiological framework for studying the gut microbiome and a means of illustrating the normal and pathophysiological structure and function of the digestive and endocrine systems. Using this model, students can be encouraged to learn the microbial identification of samples of their own oral microbiome. Additionally, information from this article could be used in a module that explains sterile technique, allows students to design experimental approaches to the study of microorganisms, communicates experimental results, and critically evaluates scientific findings related to the composition and function of the microbiome. Students might also be prompted to learn how to estimate the frequency of antibiotic-resistant cells in natural populations of bacteria and create and test their own hypothesis about patterns of antibiotic resistance. They can be encouraged to learn about the trillions of microbes living on and in their bodies and to explore the role of diet, environment, and antibiotic use in forming and maintaining individual human microbiomes, while learning to appreciate the foundations of digestive health.
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A Flavor Perception Game Designed to Introduce Basic Chemical Sensation of Taste Modalities to Undergraduate Nursing and Exercise Science Students

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Abstract
The Flavor Perception Game was designed with the goals of developing an interactive and hands-on activity, providing a platform to review chemical senses of the five tastes, promoting student investment in the course material, and providing a basis for discussion on chemical senses of the five tastes. Knowledge of taste sensation is useful for nursing and exercise sciences students, as human physiology is integral to both baccalaureate curricula. The game is inexpensive, easy to incorporate into a 50-minute lecture period, and free of chocolate allergens. Student participants (N=34) tasted three candies and completed a voluntary anonymous poll regarding their detection of the presence of umami, bitter, sweet, salty and/or sour taste modalities in the different candies. During the three stages of this classroom game, a total of 214 taste selections were made, following which students discussed various aspects of taste sensation including its importance in healthcare. https://doi.org/10.21692/haps.2019.019

Key words: active teaching methods, active learning; educational games; health professions, taste

Introduction
Human physiology is an integral course in undergraduate nursing and exercise science curricula standards. As such, student investment in physiology education is an important component in these pre-health degree programs. Pedagogical research has explored how student investment in course materials through educational interactive activities enhanced student learning and buoyed student engagement in course materials (Brown et al. 2018, Lipatova and Campolattaro 2016). One study (in a human anatomy and physiology course) identified that hands-on and interactive activities were likely to create “high-quality learning” and provide positive experiences and outcomes for participating students (Brown et al. 2017, Brown et al. 2018). Several other studies included board, word array, and educational games. These studies outlined how educational games increased student understanding of physiology topics, such as muscle cell physiology and action potentials, supported the development of study guides, and promoted student retention of new terminology (Burleson and Olimpo 2016, Luchi et al. 2017, Luchi et al. 2019, Motz et al. 2019). Another recent study reported that the use of interactive hands-on models and trivia games to complement human physiology lectures resulted in a 99% passing rate of the 152 students enrolled in an undergraduate human physiology course (Mahaffey 2018), suggesting that student engagement in activities may have resulted in improved learning through investment in course material. Additionally, a study with undergraduate dental students found that interactive activities cultivated “focused attention”, improved class participation and clarity in student cognition (Abdel Meguid and Collins 2017).

With this robust evidence of the positive attributes of including interactive hands-on components in physiology courses as a complement to lectures, the Flavor Perception Game was designed to do the following:

1. Engage undergraduate nursing and exercise students in discussions on the physiology of flavor perceptions.
2. Stimulate student discussions on the connections of physiology and healthcare using taste chemical senses as a conduit of discussion.
3. Help improve student understanding and retention of the five taste modalities and their associated chemical ligands.

This game is economical, student-friendly, examines the roles of vision and olfaction in taste sensation, and can to be played in a 50-minute lecture period, including discussion time. It examines the spiciness of cinnamon, the effects of visual and auditory priming on taste sensing for butterscotch hard candy and, finally, how odor plays a role in detecting tastes of a “mystery-flavored” candy. Additionally, the Flavor Perception Game and pre-game mini-lecture (Figure 2) target

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several HAPS Learning Outcomes for “General and Special Senses”:

- a) Identify and describe the location and structure of taste buds.
- b) Classify gustatory receptor cells based on the type of stimulus (i.e., modality).
- c) Explain the process by which tastants activate gustatory receptors.
- d) Trace the path of gustation from gustatory receptors through specific cranial nerves to various parts of the brain.
- e) Describe the primary taste sensations.

Methods

Participants and materials

The undergraduate human physiology class (N=36) to which this game was offered consisted of 11% male and 89% females students distributed as freshmen (80%), and sophomores (17%) and juniors (3%) enrolled in the undergraduate nursing and exercise science programs. Within the class, 34 students completed the taste perception polls, giving a participation rate of 94.4%. To perform the Flavor Perception Game, a number of materials common to most tasting tests, were required: 1) food source(s), 2) palate cleanser (water is recommended) and 3) survey tool. This study was approved as exempt by the Institutional Review Board of Loyola University Chicago. No IRB certification was required.

The tastants utilized in this game were candies of varied flavors. Many students verbally indicated that encountering this selection of candies was a new experience for them and it should be noted that students were not provided with nutritional information pertaining to any of the candies during the game. Nice® Brand Cinnamon Discs contained corn syrup and sugar as well as natural and artificial flavors. Nice® Brand Butterscotch Discs contained corn syrup, sugar and salt as well as natural and artificial flavors. FunDip® Valentine’s Day Mystery Flavors (purple and red packaged) were composed of dextrose, maltodextrin, citric acid, calcium stearate and natural flavors. The FunDip® candy is in a paper pouch-like packing. To open, students tear along the mid-seam of the pouch, remove the candy spatula (in one-half of the paper pouch) to scoop the powdered mystery-flavored candy substance (in the remaining half); a fun experience. The recommended palate cleanser for this game was water. Students brought their own re-usable water bottles and water stations were located outside the classrooms.

For analysis, an anonymous student poll was created in the Sakai Learning Management System (LMS) (Apereo Foundation 2018). This was done through the course web site to ensure that the polling results came from registered student participants. It is important to note the game is not limited to the selected candy or polling choices and a variety of candies (tastants) and a different LMS or survey tool can be used to execute steps of the game.

With regard to possible allergens, the candies were meticulously selected to avoid chocolate so that an allergy to that food item would not be a concern. However, the game components could not be classified as completely gluten-free. The maltodextrin and dextrin contained within the FunDip may (or may not) be derived from wheat and/or barley sources. Additionally, hard-candy molds, such as those for the butterscotch and cinnamon candies, are often coated with flour during the manufacturing processes.

Rules of play

The overall goals of the game were to have students 1) engage in an activity to help them recognize different tastes, and 2) initiate and engage in learning and critical thinking regarding taste mechanisms. The procedures were as follows (Figure 1):

1. Students were given a mini-lecture that included explaining the rules of the game and introducing the afferent taste senses and their activation. During this step students were given textbook details on the five common taste senses (sweet, salty, sour, bitter and umami) and the corresponding ligands (carbohydrates, sodium, hydrogen ion (H+), hydroxide (OH-) and L-glutamate, respectively) responsible for taste signaling (Figure 2A). These ligands were taught in accordance with the assigned lecture textbook concepts (Stanfield 2017); as such, the author recognizes that there are a myriad of compounds that would bind bitter taste gustatory receptors, in addition to the hydroxide ion.

2. Students were given one to two discs of “spicy” cinnamon candy to hold in their mouth for 30 – 180 seconds).

3. After the student had identified the taste sense (and corresponding ligand) using the web site poll, the student palate was cleansed with water.

4. Steps 2 and 3 were repeated using the butterscotch flavored candy with the following modification: the candy was described to students as “orange colored” (Figure 5).

5. Finally, students were given one packet of Valentine’s FunDip® Mystery-flavored candy (in either purple or red trimmed pouches).

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6. At this point, students were asked to hold their noses closed, to taste the candy for 30 – 180 seconds (taste in the absence of olfaction) and complete the poll.

7. Finally, students were asked to re-taste the mystery flavored candy without holding their noses closed (taste in the presence of olfaction) and, again, complete the poll.

Figure 1. Schematic of Flavor Perception Game Steps.

Figure 2. In-Class Lecture and Anonymous Voluntary Student Polling. A) Pre-game mini-lecture and instructions included mini-introductory lecture recapping afferent taste senses, gustatory taste receptors and ligands (lingual taste buds and agonists) and a number of HAPS Learning Outcomes topics. B) Results of student polling after candy sampling that were then discussed in class.
**In-class discussion**

Students began by reviewing the polling results as a class. Following the primary discussion of polling review step, secondary discussions involved student participants interviewing classmates (and inquiring of the Instructor) to help determine why some participants may have tasted flavors more intensely than others may, with questions such as:

- “Did someone have a cold (or seasonal allergies)?”
- “Was there a prior injury to the nasal area [olfactory nerves]?”
- “Perhaps previous damage to the lingual nerve [gustatory receptors]?”
- “Did a participant always – or for an extended period of time – have an aversion to certain flavors?”
- “Is the candy too sweet or spicy, for certain participants? Why?”

Finally, the Instructor also offered her own viewpoint on the candies and provided some case studies. This tri-lens (three perspectives) approach, created a variety of viewpoints from which student participants could draw conclusions on the perceived tastants (candies) and physiology of taste sensing. These discussions were carefully timed to fit within the 50 minute lecture period, which included the candy tasting steps.

**Results and Discussion**

**Cinnamon flavored candy**

Interestingly, a number of students required an additional five to seven minutes to categorize the cinnamon flavored candy, as to them “spicy” did not fall under the purview of the “five tastes”. Because of the seeming difficulty in narrowing down the “spiciness” of cinnamon flavoring, the polling results (n = 69) included all of the five taste senses. Students became invested in solving this “enigma”. Most participants (88%) selected sweet (carbohydrate complexes) as one of the taste modalities that they sensed. This aligns well with the carbohydrate content of the cinnamon candies (12 grams per serving, including eight grams of sugars). But the “other” “spicy cinnamon” flavor was more difficult to link with a single taste modality, being noted as bitter (n = 18), umami (n = 8), salty (n = 8) or sour (n = 5) (Figure 4). Interestingly, the salt (sodium) content on the nutritional packaging panel was recorded as zero grams per serving. The selection of salty by eight of 34 student game participants highlights the difficulty of underlining the cause of “spicy” cinnamon flavor detection.

Students began to ask (Figure 2B), which chemical sense was being targeted by cinnamon. This presents an opportunity for student engagement to better understand the notion of “spicy” and the links between this taste sensation and the capsaicin receptor. Furthermore, personal experience with trying to understand the perception of spiciness can consolidate

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**Figure 3. Discussion Steps. Levels of discussion: I) Personal (direct), II) Secondary (classmates and Instructor) and then III) Broader perspectives on taste – following each individual candy tasting.**

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student learning of course materials relating to taste sensation by asking students to evaluate, identify and problem solve (Brown et al. 2017, Brown et al. 2018, Mahaffey 2018). On the topic of capsaicin and spicy foods, a learning bridge connecting topics of chemical senses, nociceptors (Caterina et al. 1997) and taste can be developed in student discussion.

In 2017, a study by Trachootham and colleagues highlighted the frequency of one taste (spicy) over others in certain diet regimens, bridging the topic of physiology (taste modalities) with that of nutrition. Both are important components of the Bachelor of Science in Nursing curriculum (American Association of Colleges of Nursing 2008). Instructors can further build on the conversation related to spicy foods by introducing heat-activated ion-channels and the pain pathway, how chemical senses of taste can alert our central nervous system to the presence of an “alarming” food source, or how capsaicin receptors can in fact aggregate with sweet and/or bitter chemical sense receptors (Moon et al. 2010). The former can engage students, at this step of the game, on learning how bitter taste sensation is a peripheral nervous system function that can also protect us from poisoning (Chaudhari and Roper 2010). Students can examine how hydroxide ions (OH-), one of the major chemical ligands for bitterness flavor sense, are often a component of toxic substances and, by triggering an efferent response from the CNS to our peripheral systems of a noxious or repellent nature, can be protective against poisoning (Di Pizio et al. 2017, Reed and Knaapila 2010). The latter may help students better understand the varied game results which included bitter (OH- agonist) flavor selection (Figure 4).

Figure 4. Cinnamon-Flavored Candy Taste Senses Anonymous Student Poll. Regarding the cinnamon-flavored candy, student participants (N=34) were asked during this polling question, “Which of the following taste sensation(s) and ligand(s) were triggered?” Multiple selections were permitted.
Orange-colored butterscotch candy
An investigation into the role of visual stimulation, in regards to perceived food texture and images, was the topic of research in a recent study (van Beilen et al. 2011). During one empirical trial of this research, participants were provided with “figurative” and “non-figurative” (more abstract) image versions of strawberry and caramel items to determine the level of sweetness they associated with the visual stimuli. In this study on visual priming, participants had a perceived ranking of sweetness of food items determined by visual food textures or images from their own personal experiences with food colors, textures and corresponding sweetness (van Beilen et al. 2011). Still, it is apparent that in some cases participants in this study noted they could “see” the definitive flavor of foods presented, or predict the flavors from visual examination of the food items. A myriad of empirical investigation into the effects of the sound of music on perceived tastes has contributed significantly to our understanding of other influences on taste sensation (Höchenberger and Ohla 2018, Spence and Deroy 2013). For example, the sound of music increased the “intensity” of taste. Sweet music led to participants sensing more sweetness in the congruent samples. There were similar results for bitter food samples paired with bitter music, compared to a “no sound” control (Höchenberger and Ohla 2018).

Similar concepts presented an idea to include a section of the Flavor Perception Game that touched on the topic of interdependent sense modalities in determining properties of food or candy. Specifically, sight and sound were used, and more importantly memory and suggestion, to influence taste determination. When students were provided with Nice® Butterscotch Discs candy, the candy was described to them as “orange-colored” and students also observed the orange color of the candy. This was done to visually and auditorily “prime” students to focus on the orange color of the Nice® Butterscotch Discs. As suspected, students noted that prior to opening the candy wrapper they went from orange to surprise, due to the hearty taste expectations of their flavor perceptions and thoughts noted that upon further tasting, the immediate neural was [expecting] orange!” , “Mmmm…. “ Some students exclaimed, “It’s butterscotch!” , “I was [expecting] orange!”, “Mmmm….” Some students noted that upon further tasting, the immediate neural expectations of their flavor perceptions and thoughts went from orange to surprise, due to the hearty taste of butterscotch flavor. A few participants noted that they were likely able to sense, perhaps by smell, the butterscotch or toffee-like flavoring to negate the visual/

auditory priming toward an expectation of orange through the candy wrapper.
Student anonymous polling indicated a selection of 74 flavor votes: sweet/carbohydrate complexes = 32; salty/sodium = 23; umami/L-glutamate = 17; and bitter/hydroxide =2. There was no detection of sour (or presence of H+) in the Nice® Butterscotch Discs, noted by student participants (Figure 5). The sweet and salty selections were the top two choices and these choices were confirmed by the ingredients of the Nice® Butterscotch Discs which contained 33.3 mg sodium and 11.3 g total carbohydrate per serving. The bitter flavor was the least popular selection and possibly a guess by two student participants struggling to narrow down the unique chemical sense for butterscotch flavoring. An interesting point of note is the umami flavoring that got 17 votes. Now, another point of student engaging discussion was presented: why was the unique taste of butterscotch savory to some students?

It is interesting to note that a number of students realized that they were not fully aware of which foods can be linked to stimulation of an umami taste sensation. This presented a just-in-time opportunity to expound upon the umami gustatory response. In most human physiology textbooks (e.g. Stanfield 2017), umami is given the synonym “savory”. As such, a common suggestion for sampling is a nice steak. The suggested food items provided by the Instructor included the previous, but it is important to note that there are vegetables with naturally occurring L-glutamate (e.g. asparagus, broccoli, onions, tomatoes and a number of others) to stimulate the umami taste modality.

A number of studies on the synergism of umami tastes have been performed. Over twenty years ago, a study postulated that umami tastes receptors are activated using mechanisms similar to those of sweet and bitter taste physiological senses. For example, Fuke and Ueda (1996) examined how umami appeared to “enhance” the saltiness or sweetness of foods. In 2008, Zhang and colleagues, outlined the “functional mapping of agonist-site interaction sites” of sweet, bitter and umami tastes. It was observed that sweet and umami taste receptors “share a common subunit” (T1R3), but identify different taste stimuli. The sweet, bitter and umami tastes were all observed to be mediated by G-protein coupled receptors (Zhang et al. 2008). This illustrates another opportunity for the game to incentivize student learning and opportunities for critical cognitive processes on physiological subject matter such as the chemical sense of taste.
Due to the robust nature of butterscotch and cinnamon flavors, it is possible that the odorants released through tasting these flavors can travel to the olfactory epithelial tissue. Paul Moore notes in his book, “The Hidden Power of Smell: How Chemicals Influence Our Lives and Behavior” that somatosensory pain receptors located close to the vomer bone and associated with the trigeminal nerve can detect cinnamon odorants (Moore 2016). He further uses the previous to explain why sensing flavor differs with the ability to smell (i.e. tasting with and without the nose closed). Here an instructor can bridge the game to delve into concepts of somatosensory receptors and their role in what we perceive as taste.

Given the underlining effects of odor on flavor, the final portion of the Flavor Perception Game was designed to examine student perception of the taste of FunDip® Valentine Mystery Flavor candy, with and without odor cues. In a number of case studies with healthy subjects ranging from 18 to 80 years, participants closer to the age range of typical undergraduate students (18-20 years) exhibited higher sensitivity to taste and smells (Boyece and Shone 2006, Barragán et al. 2018), making the students involved in the current study ideal for this aspect of the game.

The cooperative sensing mechanisms of odor and taste in determining sweet flavors were explored in two studies (Stevenson et al. 1999, Djordjevic et al. 2004). The Stevenson team found that sour tastes can be “suppressed” by certain “sweet smelling” odors. In this experiment, participants indicated through rankings that the “most sweet smelling” odor of caramel was able to mask the “sourness” of citric acid solution, considered to be an excessively sour taste according to the article (Stevenson et al. 1999). The Djordjevic team investigated the odor properties of strawberry and ham on the taste of detection of sucrose products, after smelling strawberry in comparison to that of smelling ham. In this study, participants who smelled ham prior to consuming a sucrose product, tasted/detected the sweetness of sucrose at a markedly lower level of intensity than those who smelled strawberries prior to tasting the
same/similar sucrose food source. The Djordjevic team additionally highlighted studies, in which smelling strawberry, lemon, almond, caramel, maracuja and lychee also enhanced the taste of sucrose or aspartame food products (Djordjevic et al. 2004).

In this portion of the Flavor Perception Game, students noted that without an odor cue there was minimal detection of the full spectrum of flavors in the mystery flavored candy. Once students were able to use odor cues and smell the candy, a more robust spectrum of flavors was detected. Students were encouraged to discuss how the olfactory senses (food odors) may affect the sense of taste and whether the intensity of taste is similar when one has a "stuffy" or injured nose. Students found the topics of colds (rhinovirus) and other effects on smelling compelling. It was noted that food does not taste as good when you have a stuffy nose. This presents the question of whether or not the olfactory nerves have a function in the gustatory system. During this game, student participants may also be inclined to discuss how the intensity of flavor perception declines in patients with nose injuries or physiological disorders affecting smell. For the undergraduate health professions student participants, this game offered discussion topics that may be useful in their future patient care experiences, helping them to comprehend the difficulties faced by patients with impaired olfactory senses.

The results of the FunDip® Valentine Mystery Flavor candy anonymous student poll (n = 71 total flavor votes) presented nearly equal votes for sweet (n = 28) and sour (n = 32) (Figure 6). These results help to explain the seemingly split vote between citric mystery flavor guesses (51% orange or lemon) and berry or watermelon (49% voted; n = 57) (Figure 7), among student participants. Bitter flavor was detected at a lower frequency (n = 11). It is likely, as most students discussed, that the bitter flavor was noted in "hints" of the presumed "citric" tastes. The mystery flavored candy, per nutritional panel information, contained 11g of total carbohydrates (which included the total 11g of sugars) and zero mg of sodium for a one packet serving, with less than 2% citric acid. The sodium and sugar content of the mystery flavored candy supports the high sweetness taste detection and no salty detection of the candy. The citric acid (hydrogen ion donor) ingredients, although less than 2%, likely accounted for the sour taste.

**Figure 6.** (Mystery Flavored) Valentine’s Day Candy Taste Senses Anonymous Student Poll. Regarding the Valentine’s Day (mystery-flavored) candy, student participants (N=34) were asked during this polling question, “Which of the following taste sensation(s) and ligand(s) were triggered?” Multiple selections were allowed.
Concluding Remarks
During the game, students commented positively on this fun approach to learning the five taste senses, gustatory cells receptors, and ligands. Participants described how they enjoyed tasting the samples and examining how taste depends on selective ligand-receptor interactions. Some students noted that the visual cues (colors) of the candies caused them to “expect” a different flavor (as in the butterscotch) than the one they experienced upon tasting. The mystery-flavored candy was a sensational hit. Students responded well to this confectionary conundrum and took samples home to further examine the mystery flavor. Lastly, it was stated that the game offered student participants an opportunity for them to perform an in-depth analysis of candy products, review the ubiquitous nature of gustatory ligands in candy products, and examine how the olfactory, auditory, vision and taste neural pathways work in a cooperative effort in perceiving the five types of tastes.

In conclusion, the Taste Perception Game served as a platform to promote critical thinking, student discussion, investment in course material, and opportunities to bridge a topic of human physiology and healthcare.

Acknowledgements
The author would like to acknowledge the student participants of this game.

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Figure 7. Determine the Mystery Flavor of the Valentine's Day Candy Anonymous Student Poll. Student participants (N=34) were polled and instructed to select up to TWO possible detected flavors of the mystery-flavored Valentine's Day Candy.
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continued on next page
A Flavor Perception Game Designed to Introduce Basic Chemical Sensation of Taste Modalities to Undergraduate Nursing and Exercise Science Students


MASTER OF SCIENCE IN HUMAN ANATOMY AND PHYSIOLOGY INSTRUCTION

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Method For Production of 3D interactive Models Using Photogrammetry For Use in Human Anatomy Education

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Abstract
Despite a shift to use digital resources to supplement traditional anatomy education, institutions often rely upon external sources of digital materials. Such three-dimensional (3D) animations rarely resemble the anatomical models or cadaveric specimens used in the students' laboratories. Photogrammetry is a technique that generates an interactive three-dimensional model from a series of photographs. This study developed a simple and inexpensive method for using photogrammetry to produce interactive models that can be used by the anatomy educator. Only cell phone cameras were used, and the authors had no previous experience with photogrammetry. Such photo-realistic interactive models of cadaveric specimens or plastic anatomical models may allow learners to review and recall anatomical structures seen in the laboratory on any web-enabled device.

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Key words: anatomy education, digital anatomy, photogrammetry

Introduction
Undergraduate human anatomy courses are taught in a variety of educational institutions from two-year community colleges, to four-year colleges, to universities. A wide variety of instructional practices are common, including cadaveric dissection and prosection, physical models, and clay modeling (Lombardi et al. 2014; Motoike et al. 2009; Waters et al. 2005). Digital models are used to supplement or replace more established pedagogical tools (Fredieu et al. 2015; Saltarelli et al. 2014), and virtual reality and 3D printing are the newest additions to the toolkit of anatomy educators (Fredieu et al. 2015; McMenamin et al. 2014). From this great diversity of visualization tools, each institution makes choices based on pedagogical rationale, budgetary concerns, the educational level of the students, and/or the level of experience and expertise of those in the position to make such decisions.

Anatomy students may encounter major barriers to outside-of-class access to the same materials used in their laboratory instruction. Whether students are unable to access these materials because there are too many of them vying for laboratory time or their own busy academic and extracurricular schedules do not afford the time, limited access to the educational resources used in their laboratories can negatively impact their learning. Digital products, with their ubiquity in today's world, have entered to fill this void. Such applications allow students to learn or review anatomy on their own time and in any place. Sugand et al. (2010, p. 85) argued that “the future of anatomy teaching must rely more on visual aids outside the dissection room.”

For many students, however, the available cadaver dissection software or 3D model may bear little resemblance to the materials used in their college or university’s laboratory. Some colleges may use prosected cadaveric specimens in the laboratory but provide students with 3D models that bear more resemblance to cartoons than to their laboratory materials. Bridging the gap between the physical materials in the laboratory (e.g., model, prosected cadaver, or organ specimen) and a software package of idealized anatomy may be difficult, particularly for the novice learners of anatomy often found in undergraduate classrooms. These students would likely benefit from digitized images of their actual physical laboratory materials. However, 2D images may be difficult to use because they do not portray the spatial relationships visualized on a 3D model or specimen.

Photogrammetry is a tool that utilizes multiple photographs to generate a complex 3D surface model (Luhmann et al. 2006). Available software identifies points on a photograph, and matches them to similar points taken from a slightly different angle. With dozens of photographs, aligned points are used to generate a 3D representation of these points, a point cloud. From the point cloud, polygons are added to create a surface mesh. A texture is applied to the mesh, wherein the original photographs are “stitched” together to form a complete image for each of the polygons. Unlike surface scanning, photogrammetry does not use expensive laser scanners; instead modern mobile phone cameras are sufficient to take photographs. In addition,
rather than simply provide a surface reconstruction, photogrammetry compilation software applies the color and simulated texture of the object. The result is a photorealistic 3D model. Previously, this technique has been applied to modeling pathology specimens (Turchini et al. 2018), and to isolated human organ specimens (Petriceks et al. 2018). These studies required expensive rendering software and cameras; the latter utilized a proprietary multi-camera and lighting setup developed by Anatomage, Inc.

The purpose of this study was to develop a simple protocol to inexpensively create interactive 3D digital models of anatomical learning materials. This novel tool in anatomy education leverages the ubiquity of a web-based platform such as Sketchfab for viewing 3D models and virtual reality and allows students to see the very same cadaveric specimens or plastic models found in their laboratory. Importantly, we demonstrate that this approach can be used with very little financial commitment and with very little prior technical expertise, so it can benefit anatomy educators in institutions of any size.

Materials and Methods
To test the proof of concept that photogrammetry could generate accurate 3D models for use in anatomy education, two teaching resources were used: a cadaveric hand and forearm (used to demonstrate the muscles and tendons of the hand and wrist), and a model of the upper limb (3B Scientific, Germany). Figure 1 illustrates the workflow for producing the 3D models.

Specimen Preparation and Photography
Preparation for photography was minimal. A typical, unaltered teaching laboratory served as the location for photographs. Reflective surfaces in the room can create difficulty in the alignment process used to create 3-D images. It is recommended that mirrors or other reflective surfaces, such as a glossy whiteboard, should be covered or obscured. Additionally, the authors observed that an uninterrupted, expansive background of a single color that closely matches the color of the subject of photography might result in fewer matches in point alignment, so this was avoided. In the case of the cadaveric specimen, wet locations were blotted dry prior to photography to reduce the likelihood of reflection.

In positioning the specimen, it was desirable to have as much access around the specimen as possible in order to enable photography from a variety of perspectives. No lighting apparatus or backdrops were necessary to produce models of sufficient quality. Ambient laboratory lighting was sufficient, provided no distinct shadows were present on the object of photography. In preliminary trials, a mobile phone ring light (www.flawlesslighting.com) was used, but was found to be unnecessary. If a space with too much directional lighting cannot be avoided, a ring light might prove useful.

Figure 1. Workflow illustrating the generation of 3D models.
Photographs were taken using a 2016 iPhone 7 (Apple, www.apple.com). The auto-adjustment of the native camera application’s ISO (International Organization of Standardization – assesses the sensitivity of the image sensor) value resulted in highly variable photographs with regard to brightness. Depending upon which objects were present in the background with a particular camera angle (a black benchtop, a bright fluorescent light), the auto-adjustment of ISO affected the appearance of the object. Thus, the object’s brightness was inconsistent when compared across photographs. Locking the ISO at a consistent value resulted in images of the object that were consistent in brightness regardless of the background. Subsequent photography was conducted using the Moment app (www.shopmoment.com), which allowed for the locking of ISO.

At least 100-200 photographs were taken, encircling the model as well as at multiple angles to capture the model from many visual perspectives. Figure 2 illustrates the calculated angles from which 167 photographs of the model arm were captured. No precise regimen of determining angles was necessary. The authors simply took small steps to encircle the object, taking 20-30 photographs; then, a higher or lower camera perspective was taken and another 20-30 photographs were taken encircling the object. In total, the authors found that four to six different angles with respect to the z-axis along with an approach to encircle the object at each of these angles provided sufficiently accurate and reproducible results. The photos were taken with an attempt to either fill the photograph with the entire region of interest or with a slightly closer approach. In this study, the distance was approximately 30 cm. Photographs were 3024×4032 (12 megapixels); camera settings were 0.033 second exposure; ISO speed = 32; f 1.8. The photography of a single specimen took approximately 5 to 10 minutes. The resulting collection of digital photos was screened, and out-of-focus photos were deleted. Approximately 100 to 200 photos proved sufficient to produce high-quality 3D models.

Photogrammetry
Agisoft Metashape standard edition, version 1.5.0 (www.agisoft.com; subsequently referred to as Metashape) was used to compile the photographs, generate a point model, and produce the mesh model and its texture. This software is available for Microsoft Windows, Linux, and macOS. Other software is available at no cost, for example: Photomodeler (www.photomodeler.com), Meshroom (alicevision.org) and RECAP (www.autodesk.com). The authors found Metashape to have no compatibility issues with their laptop computers (Apple, www.apple.com), easy to use, and the one-time standard, educational license was $59 at the time of this study. Tutorials are available on the Metashape web site (www.agisoft.com/support/tutorials/beginner-level/). After importing the approximately 100-200 photos into Metashape, they were aligned using the high accuracy setting to generate a sparse point cloud (Figure 2). The default selections were unmodified (generic preselection; key point limit = 0, unlimited; tie point limit = 1000; adaptive camera model fitting) for this step.

Figure 2. Sparse point cloud generated from 167 photographs of an anatomical model in Metashape (www.agisoft.com). Matching points in photographs are compared to calculate the position of the camera in space (blue rectangles), and a 3D representation of the mapped points is generated. An effort was made by the authors to methodically encircle the model at a variety of perspectives, as can be seen by the illustrated camera positions. Left and right panels show the same sparse point cloud from two perspectives.

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The sparse point cloud that was generated was then edited to remove points not related to the object. Options for reducing error were performed: reconstruction uncertainty (value chosen: 10), projection accuracy (value chosen: 10), and reproduction error (value chosen: 1). The photos were then realigned with the remaining points and the region of interest was defined by resizing the bounding box to include only the specimen. A medium accuracy dense point cloud was then generated (Figure 3A). Processing time for higher accuracy density point cloud generation was significantly higher and produced results that did not justify the additional processing time. The resulting dense point cloud was manually edited to remove any erroneously identified points, not part of the anatomical specimen or model. A mesh was generated from the points with a target of 1,000,000 faces (Figure 3B). Meshes with many more faces produced files that were too large, and the added detail was not relevant for this application. Other settings were left as default (Surface type = arbitrary 3D; Interpolation = enabled; Calculate vertex colors). A texture was applied to the mesh using the default settings (Mapping = generic; Blending = average; Texture size/count = 4096 x 1; Hole filling = enabled).

The resulting model (Figure 3C) was exported in OBJ, JPG, and MTL file formats. The OBJ file contains the mesh, the JPG and MTL files contain the texture and its properties. Decimation of the resulting mesh was not performed since the mesh was originally created with a 1,000,000 face target, but excessively large models can be slightly simplified within Metashape or other meshing software without obvious effect on model quality. Models with 100,000 to 1,000,000 faces were determined to be of excellent quality for the proposed application.

**Figure 3.** Refinement of the model. (A) Dense point cloud is manually edited by selecting erroneous points with mouse cursor. Points that are obviously not a part of the original object are evident when rotating the dense point cloud. The authors found it unnecessary to remove erroneous points internal to the model. (B) A 1,000,000 face mesh is produced from the dense point cloud. Metashape assigns a single color to each polygonal face, but only is a more photorealistic model produced when (C) a texture is applied; each face of the mesh receives an image derived from the original photographs.
Archival and retrieval
OBJ, MTL, and JPG files were imported into the web service, Sketchfab (www.sketchfab.com), which provides a “Pro” account for educators at no cost. Within Sketchfab, 3D objects were manipulated to remove the defaults for lighting, position, and other aesthetic factors. If desired, these variables can be adjusted to the instructor’s preference. Additionally, labels can be attached to the model from within the Sketchfab web interface. An instructor-provided URL was generated to enable student access to interactive 3D models in Sketchfab. URLs could be provided directly to students or, alternatively, links to the models could be embedded in an HTML-based webpage using an embed code generated by Sketchfab. The 3D models were viewable on any web-enabled device, regardless of platform, and virtual reality could also be used. Additionally, download of the file for 3D printing could be enabled. A website was developed for students, allowing easy access to all models made using the same methods: https://anatomy.web.unc.edu

Student Perceptions of Usefulness
An anonymous end-of-semester evaluation was included as a component of a larger course and instructor evaluation. Students were asked to evaluate the usefulness of various provided or linked resources within the course management system. In addition to the 3D virtual models described in this paper, the items to be evaluated included: a custom laboratory text, a slide deck that was also used in each laboratory lecture period, online flashcards customized to the content of the laboratory, and various online videos, websites, or mobile apps deemed to be relevant to the content of the laboratory. Suggested mobile apps were free of charge. With the exception of the lecture slides, all of the resources were presented as materials for students to study outside of laboratory and not used directly in laboratory instruction.

Results
The methods employed in this study successfully produced 3D digital models of a plastic anatomical model and of a cadaveric prosection. Screenshot images of the completed models are shown in Figure 4, and the interactive 3D models can be accessed with the following links: arm dissection: https://sketchfab.com/3d-models/ac0b6ada828349b99d4e9fa6a9cc4fbd; arm model: https://sketchfab.com/3d-models/1d75fd9232144f1b9467b6d2d1639b48. The interactive 3D models can be rotated in three axes, scaled up or down, and moved in a planar dimension. Students were provided with access to the models through links in their course management system, so they could use them for review of structures seen in the laboratory.

Figure 4.
Screenshots of final 3D interactive models.
The level of detail provided excellent visualization for the purpose of learning and reviewing anatomical structures inside or outside of the laboratory. The cost of producing interactive 3D models using photogrammetry may differ according to existing supplies. However, for the current study and using materials that were previously purchased for common computing tasks, the only monetary cost of producing the interactive models was the one-time purchase of the photogrammetry software.

End-of-semester evaluations revealed that 73.6% of 413 respondents found the 3D virtual models to be either very useful or somewhat useful resources (Table 1). A small number of respondents found them unhelpful or very unhelpful, numbers commensurate with those of the other resources. The laboratory text was more highly rated, but all other resources did not achieve the same usefulness ratings as the 3D virtual models.

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<th>N/A, didn’t use</th>
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<th>Somewhat helpful</th>
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Table 1. Summary of end-of-semester student evaluation of lab resources. Values indicate percentage of 413 anonymous evaluations of the usefulness of provided resources for a 1 semester undergraduate anatomy laboratory.

**Discussion**

The aim of this project was to develop a simple methodology for the production of interactive 3D models of laboratory materials. We have demonstrated that accurate, interactive 3D models of anatomical teaching specimens can be produced for the benefit of students in undergraduate anatomy courses at two-year community colleges, four-year colleges or universities. Using inexpensive software and common laptop computers and mobile phones, instructors with no photogrammetry experience will be able to produce realistic digital models that are accurate reproductions of laboratory models, prosected cadaveric materials, human or animal organs, or even regional dissections of a cadaver.

Apart from the ability of students to review a virtual replica of their laboratory models or specimens, the interactive models also have potential use in instruction. The web platform described allows users to easily generate a code to embed the interactive 3D model in HTML-based instructional materials. The authors have opted to leave their virtual models unlabeled, for the purpose of post laboratory review, however instructors may label their models with terminology, origins/insertions/actions of muscles, or simply number anatomical structures for short formative assessments. Such instructional and assessment tools would be available to students on any device capable of rendering
Method For Production of 3D interactive Models Using Photogrammetry For Use in Human Anatomy Education

HTML. Alternatively, the generated OBJ files can be inserted into a PDF document format capable of rendering 3D for distribution to students.

Student evaluations indicate that respondents generally found the 3D virtual models useful. The laboratory text had the highest rating of usefulness, likely as a result of its central role in defining the course’s content. Among the other resources, the 3D virtual models were clearly valued by most students in their out-of-classroom study. This novel learning tool allows undergraduate or medical anatomy students to have access to the exact laboratory specimens or anatomical models that they use in the laboratory. Thus, with the development of photogrammetry-based interactive digital 3D models, students out of the classroom have additional tools to support their success in anatomy courses with minimal effort from course instructors.

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Zachary J. Burk is a recent graduate of the University of North Carolina, with a BS in Biology. He is an Internet entrepreneur and he has been teaching human anatomy and physiology for two years. Zach is currently a first year DDS student in University of North Carolina’s School of Dentistry.

Corey S. Johnson, PhD, is a Teaching Associate Professor and Associate Chair of Biology at University of North Carolina. His interests are human and vertebrate morphology, physiology, and development.

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Back to TOC
“Stringing Together” Capillary Exchange

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Abstract
In Anatomy and Physiology courses, students often struggle with visualizing structures, or processes, that occur at the cellular level, such as capillary exchange. Whenever available, models can be vital to students’ understanding of a topic, particularly when they can be constructed by students in real time. To demonstrate capillary exchange to freshmen and sophomore Anatomy and Physiology students, a senior Health Sciences student created a simple model. The student enrolled in an independent study as a near-peer laboratory assistant. In addition to assisting the Anatomy and Physiology students learn in the lab, the near-peer student was asked to create a tool for active learning of a difficult physiological concept. Presented here are the materials and steps to create the model, recommended exercises for students to complete with the model, and example assessments that demonstrate how the activity helps students meet the relevant HAPS learning outcomes.

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Key words: anatomy, physiology, capillary, models, active learning

Introduction
There are many structures and processes in Anatomy and Physiology that students find challenging to visualize and understand, particularly when concepts traditionally learned in biology, chemistry, physics or mathematics courses are involved. When these structures or processes are not easily visualized, students can experience even greater barriers to learning (Crowther 2017; Lujan et al. 2013; Rodenbaugh et al. 2012). One such process is capillary exchange. In Anatomy and Physiology courses, this topic is often introduced when studying the cardiovascular system, yet the principles are introduced very early in the Anatomy and Physiology curriculum when discussing diffusion and osmosis.

Hands-on exercises such as graphing, drawing and modeling can help students overcome barriers to learning (Crowther 2017; Hull 2016, Motz et al. 2017). In particular, models have frequently been well-received by students in Anatomy and Physiology courses, and have great potential to demonstrate structures and processes that are difficult to visualize (Breckler and Yu 2011; Dirks-Naylor 2016; Hull 2016; Motz et al. 2017). To help students visualize capillary exchange, a simple model was created by a near-peer laboratory assistant to meet the requirements of an Honors Project. The student was enrolled in a three-credit independent study, and was present at every weekly Anatomy and Physiology laboratory session. The student was eager to create an inexpensive, easy to understand model that students could use.

Materials
Only a few simple, inexpensive materials are needed for the representation of blood capillary exchange:

1. Red and blue string (purple string can be added to demonstrate the transition from artery to vein.)
2. Construction paper of various colors
3. Scissors

The instructor should provide the materials and instructions, but allow the students as much freedom as possible to create their own representation of the capillary system. This model can be created individually or in small groups in laboratory, recitation or lecture setting, as long as tables are accessible.

Methods
In this assignment, students construct a visual representation of a capillary, with the goal of understanding how the various forces on the capillary wall influence exchange of materials between the blood and interstitial fluid. It is recommended to start by laying out the strands of string, using red and blue colors to represent the artery and veins. Additionally, purple strands can be incorporated to show the change from oxygenated red arteries to deoxygenated blue veins through the capillary. If any students have color vision limitations, strings with different thickness (thicker for arterial end, thinner for venous and thinnest for capillary, should be on hand.
After creating the capillary, students will create their own models of each molecule that passes through the capillary system. Key molecules to create are O₂, CO₂, H₂O, albumin, platelets, red blood cells, and white blood cells. These molecules can be cut in whatever shapes the students feel most comfortable with; however, it is important to emphasize the size difference in molecules to later aid in demonstrating why some molecules are able to move in and out of the capillary.

As students build their own models, it may be helpful to provide images from the assigned textbook or learning resource to students having difficulty getting started, or to allow students to check a reference image from time to time. If time is limited, instructors may want to circulate between students or student groups to ask questions about the students’ models, and ensure each part is critiqued if necessary. A recent study by Dirks-Naylor on the student construction of nephron models to facilitate understanding of renal transporters found that students prefer confirmation from the instructor that the model was drawn correctly to increase their confidence (Dirks-Naylor 2016). If more time is allotted, students should be allowed to make mistakes in construction and rebuild, as this can be an excellent learning opportunity. In their model of the sarcomere, Rodenbaugh and colleagues stressed the importance of allotting sufficient time during active learning exercises for thinking and processing, if life-long learning is to occur (Rodenbaugh et al. 2012). Indeed, allowing students the freedom to deconstruct, reconstruct and manipulate the capillary model based on their own independent theories and conclusions puts the responsibility of learning on the student, rather than the instructor. While the length of the model and diameter of the vessels can vary, it is
important to make sure that students are accurate in differentiating the sizes of certain molecules. Water and oxygen molecules should be small enough to move through the holes built in the capillary, and albumin molecules should be too large to fit through these holes.

Once the students have created all their own materials it is important to make sure they are able to explain the basics of blood capillary exchange. Emphasis should be on how hydrostatic pressures and osmotic pressures affect the movement of fluid and therefore the molecules with them. When students can confidently create and explain their own models of the blood capillary system, instructors can give each group a physiological example of how the capillary system can change, causing different movement of molecules and fluid. For example, instructors can ask students to demonstrate how high or low blood pressure, or low albumin levels, could change the exchange of molecules across their capillary model. Once the groups have been given time to model their own capillary response, it is important to than have each group take turns explaining to other groups, to allow students to learn from each other. As students begin to demonstrate a clear understanding of their models and the capillary system, there are a variety of ways the instructors can incorporate further details and information. Simple additions could include: incorporating additional strings or other material to demonstrate the different layers of the blood vessels; adding more molecules found in the blood plasma; and focusing on the different roles that ions, proteins, and cells play within the blood vessels and capillary system.

Student Activity Instructions
An example student instruction page for the activity is provided below.

Example of in Class Instruction

**Part 1**
In small groups, use the provided material to create a representation of the blood capillary and associated molecules to demonstrate net filtration. Include the following molecules:

- Water
- Oxygen
- Carbon Dioxide
- Red Blood Cells
- White Blood Cells
- Albumin
- Platelets
- Glucose

Once you have created your own model, notify your instructor. Demonstrate your understanding of how hydrostatic pressures and osmotic pressures affect the movement of molecules in and out of the capillary.

**Part 2**
Once your instructor has approved of your model, they will provide you with one of the following physiological events, and task you with demonstrating how it affects the capillary system:

1. Please explain and demonstrate how histamine release in the interstitial fluid will cause changes in the capillary system. Additionally, please provide an example of what will cause this histamine release to occur.

2. Please explain and demonstrate how edema can be caused and how the lymphatic and capillary systems are directly related in preventing this condition.

3. Please explain and demonstrate how low blood pressure will lead to changes in the capillary system and how the body must adapt to maintain homeostasis.

Once everyone in class has finished creating their own model, groups will take turns demonstrating and explaining their physiological event to the class.
Assessment
Grading of this assignment can be based on completion of the required tasks, participation, or on demonstrated understanding of the material. Specifically, for grading options, students’ grades can be determined by their ability to demonstrate and clearly explain a physiological response that occurs in the capillary to the rest of the class. Students could also make a short video that can be uploaded to a learning management system, and graded by the instructor or peers.

Through this activity, students are able to achieve multiple HAPS learning outcomes. Students are able to comprehend how the composition of capillary walls differs from that of other blood vessels. They are able to correlate the anatomical structure of capillaries with their function through building and explaining the basics of the capillary system (HAPS module K, topic 12, learning outcome 4a and 4c). Creating and demonstrating the gaps in the walls of capillaries as well as using different string thickness for each blood vessel type allow students to associate the different key characteristics of each blood vessel region. Creating molecules and explaining how these molecules move across capillary walls allows students to understand the role each molecule has in the process of the exchange of gases, nutrients, and wastes across capillary walls (HAPS module K, topic 14, learning outcome 7a). Having students explain the process to professors or teaching assistants allows instructors to ensure that students comprehend the roles of filtration and reabsorption in capillary exchange and how net filtration pressure across the capillary wall determines movement of fluid across the capillary wall (HAPS module K, topic 14, learning outcome 7b, 7c, and 7d). Lastly the incorporation of specific physiological conditions will provide students with examples of how the cardiovascular system and capillaries respond to environmental changes to maintain homeostasis in the body (HAPS module K, topic 15, learning outcome 1).

Depending on available time and goals for the session, instructors can take this model even further in demonstrating hydrostatic pressure and colloid osmotic pressure. Instructors could provide the equation for net filtration pressure (NFP), give students sample values, and ask students to make a calculation, using the model to demonstrate their understanding of the correct equation answer. Allowing students to move the molecules while simultaneously discussing the factors that determine NFP could help students gain greater confidence and understanding in the relationship of these equations.

Student Feedback on Model
As the model was created as an Honors Project for the near-peer student, the model was not created in time to implement it in an actual laboratory or classroom setting in Anatomy and Physiology 103, the final course in the three-course sequence. However, the near-peer’s mentor, the Director of the Anatomy and Physiology Curriculum, held a final exam review session for students.

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Grading Rubric

<table>
<thead>
<tr>
<th>Part 1</th>
<th>Score</th>
<th>Criteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Students ability to design and</td>
<td></td>
<td>1. Student met the minimum required</td>
</tr>
<tr>
<td>model the blood capillary</td>
<td></td>
<td>2. Student met expectations</td>
</tr>
<tr>
<td>system</td>
<td></td>
<td>3. Student clearly exceeded expectations</td>
</tr>
<tr>
<td>Students ability to understand</td>
<td></td>
<td></td>
</tr>
<tr>
<td>and explain the blood Capillary</td>
<td></td>
<td></td>
</tr>
<tr>
<td>system</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Total Score: ___/12

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Figure 3: Example rubric for assessment of the activity

Figure 4: Near-peer student and completed model for demonstration
During the exam review, the near-peer student demonstrated the model to a group of ten students and described the proposed activity to them as described in this paper. Every student agreed that this would have been an excellent activity in lab, small groups during lecture, or for the lecture instructor to demonstrate capillary exchange in class. One student commented “I wish I saw this before the midterm!” which was when the topic was first introduced. She reported being very focused on memorizing the values for net filtration pressure at the arterial and venous end, but struggled to understand how those pressures were created. After demonstrating the model, each student was able to move the molecules in the correct directions under normal conditions, and then in the proposed physiological conditions in the activity: low albumin, histamine release, and low blood pressure. When the near-peer student attached the histamine molecule to the capillary and widened the gaps between the strings, a visible “aha” moment occurred in the group. The students were able to see how white blood cells could reach interstitial tissue. Next year, the activity will be included in the laboratory that focuses on the heart and blood vessels.

Limitations and Future Directions
A simple model using only string and paper can be used to demonstrate the basic forces influencing flow across the capillary wall. Once students have a solid understanding of what is exchanged across the capillary, and why, they can better apply the equations to calculate net filtration pressure. Models have the potential to be even more useful for students when created by former students who were successful in their Anatomy and Physiology courses. Broader implementation of the model is necessary to fully demonstrate the effectiveness of the model as a teaching and active learning tool. While students may be excited and engaged by the activity, that does not necessarily mean that the activity achieves higher-order thinking or understanding of a concept, which is a primarily goal of active learning strategies. Motz and colleagues’ soda bottle model of a nephron was met with great student enthusiasm, and helped students understand glomerular filtration, but still required instructor explanation for the forces behind reabsorption and secretion in the tubule (Motz et al. 2017). Instructors may assume that because students give positive feedback about a model, it is helping them better meet the desired learning outcomes. A 2017 literature review of active learning strategies by Hopper found that the success of active learning strategies is variable, and recommends instructors critically evaluate the effectiveness of their classroom activities with an evidence-based approach (Hopper 2017).

It is also important to recognize that very simple educational models such as the one presented here have limitations. For example, no fluid can “flow” through a capillary to demonstrate the real-time effects of varied hydrostatic and osmotic pressures in the capillary and surrounding extracellular fluid. However, this model could be an excellent starting point, particularly for students with no prior knowledge or exposure to the information. Instructors could then provide students with more detailed illustrations, or videos, demonstrating capillary exchange. There are a great number of potential clinical applications students can use the model to demonstrate, many of which they would routinely see in clinical practice. Finally, the Anatomy and Physiology students in the laboratories with near-peers regularly expressed excitement and awe that the near-peers had been in their shoes a year prior. Therefore, models may be even more beneficial to students when developed by their near-peers who were successful in the course, as they are closer to them in terms of knowledge and experience.

About the Authors
Alejandro Quinonez, BS, is a recent graduate of the Drexel University Bachelor of Health Sciences Program. In the spring of 2019 he served as a near-peer assistant in the Anatomy and Physiology laboratories.

Krista Rompolski, PhD, is an Associate Professor in the Health Sciences Department at Drexel University. She supervised the near-peer students in the Anatomy and Physiology laboratories for the 2018-19 school year.

Literature Cited


Motz VA, Suniga RG, Connour JR. 2018. Inexpensive hands-on activities to reinforce basic physiological principles: details of a soda bottle nephron model. HAPS Educator. 22(2): 159-164.

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<td><strong>AWARDS &amp; SCHOLARSHIPS</strong></td>
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<td>Mark Nielsen</td>
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