Cognitive and non-cognitive outcomes associated with student engagement in a novel brain mapping and connectomics course-based undergraduate research experience

Christina E. D'Arcy[1,3,5–7]*, Anais Martinez[1–3,6], Arshad M. Khan[1,3,4,6,7]‡, and Jeffrey T. Olimpo[1,4,5,7]‡

[1]Department of Biological Sciences, [2]Doctoral Program in Pathobiology, [3]UTEP Systems Neuroscience Laboratory, [4]Border Biomedical Research Center, [5]Biology Education Research Group, [6]HHMI PERSIST Program, and [7]NIH BUILDing SCHOLARS Program, University of Texas at El Paso, El Paso, TX 79968, USA.

‡ co-senior authors

**Address correspondence to*: Christina E. D'Arcy, Ph.D., Postdoctoral Research Associate, Department of Biological Sciences, University of Texas at El Paso, El Paso, TX 79968. Tel: +1 (915) 747-8825. E-mail: cebond@utep.edu

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Author contributions

AMK, AM, CED developed the course curriculum and pedagogical design. AMK provided the course project and protocol design. CED, AM, JO, and AMK developed the map evaluation rubric. CED and AM provided map evaluations. Course evaluation approach and design of the exit survey were provided by JTO and CED. Statistical analysis was performed by JTO and CED. All authors contributed to the writing and evaluation of the manuscript content.

Abstract

Course-based undergraduate research experiences (CUREs) provide an innovative platform for engaging emergent scholars in the authentic process of scientific discovery. Current evidence indicates that CUREs are effective at fostering students' development of content knowledge, motivation, and persistence in the science, technology, engineering, and mathematics (STEM) disciplines. Importantly, CUREs simultaneously offer investigators unique access to an extended population of students who receive education and mentoring in conducting scientifically-relevant investigations and who are thus able to contribute effort toward big-data projects. While this paradigm benefits fields in neuroscience, such as atlas-based brain mapping of nerve cells at the tissue level, there are few documented cases of CUREs in the domain.

Here, we describe a curriculum designed to address this deficit, evaluate the scientific merit of student-produced brain atlas maps of immunohistochemically-identified nerve cell populations for the rat brain, and assess shifts in science identity, science communication, and experimental design skills of students engaged in the introductory-level Brain Mapping and Connectomics (BM&C) CURE. BM&C students reported gains in research and science process skills following participation in the course. Furthermore, BM&C students experienced a greater sense of science identity, including a greater likelihood to discuss course activities with non-class members compared to their non-CURE counterparts. Importantly, evaluation of student-generated brain atlas maps indicated that the course enabled students to produce scientifically-valid products. Together, these findings support the efficacy of the BM&C course in addressing the relatively esoteric demands of chemoarchitectural brain mapping.

1. Introduction

Course-based undergraduate research experiences (CUREs) are broadly recognized as an effective active-learning approach in a laboratory context (Chickering & Gamson, 1987; Bonwell & Eison, 1991; National Research Council, 1996; 2003; Handelsman et al., 2004; American Association for the Advancement of Science, 2011; PCAST, 2012; Auchincloss et al., 2014). Several key aspects distinguish CUREs from other active-learning approaches and undergraduate research experiences. Briefly, through a collaborative and iterative process, CUREs engage students in scientific practices that promote discovery of novel findings that are of broader relevance to the scientific community (Auchincloss et al., 2014). The resulting shifts in course enjoyment, research skills development, autonomy, and retention observed for CURE students versus their non-CURE counterparts have motivated interest in developing CURE curricula across the STEM fields (Badalà et al., 2013; Auchincloss et al., 2014; Jeffery et al., 2016; Olimpo et al., 2016; Rodenbusch et al., 2016; Ballen et al., 2017; Dolan, 2017; Frantz et al., 2017). In neuroscience, while problem- and research-based courses within the subdisciplines of medicine (medical neuroanatomy), psychology, and cell/molecular biology are represented in the literature, a brief review of published curricula indicates a gap in CURE offerings designed for entry-level students (please refer to **Supplemental Materials I** for a description of such approaches). Moreover, none of the curricula reviewed approached big-data problems in neuroscience, nor engaged students in atlas-based neuroanatomical mapping. The limited presence of such CUREs may be attributable to a prevailing assumption that such nuanced work requires extensive knowledge of neuroanatomy that can only be mastered through upper-division coursework. Yet, if lower-division CUREs could be designed to successfully incorporate laboratory-based curricula that involve students acquiring basic atlas-based neuroanatomical mapping expertise, the advantages afforded to the research community by effectively crowd-training this labor-intensive process would be significant.

We deliberately use the term "crowd-training" instead of "crowdsourcing" to acknowledge that a training component is required before individuals could perform the specific type of neuroanatomical mapping techniques we describe here. Given that crowd-trained individuals (students in CUREs) have helped advance big-data projects in the field of genome annotation (Chen et al., 2005; Call et al., 2007; Elgin et al., 2016), and that certain projects within the sub-domains of pathology (Della Mea et al., 2014) and neuroanatomy (Roskams & Popović, 2016; Irshad et al., 2017; also see Oleson, 2011) have been crowdsourced successfully outside of a formal CURE environment, we reasoned that it was possible to develop, implement, and evaluate a CURE that provided students the means to conduct authentic scientific research in atlas-based neuroanatomical mapping.

Accordingly, with the support of a grant from the Howard Hughes Medical Institute (HHMI) (Grant 52008125), we created an introductory biology CURE called *Brain Mapping & Connectomics (BM&C)*, the first CURE of its kind in the country. Recently concluding its fifth year (Martinez et al., 2019), the research goals for students in BM&C are to 1) use histological methods to label and identify the chemical phenotypes of hypothalamic neurons in brain tissue; and 2) to generate high-spatial-resolution chemoarchitectural maps of these neurons and their axonal projections using a standardized reference atlas of the brain (Swanson, 2004). Through this process, the students make discoveries concerning novel patterns of distribution and relationships among neuronal phenotypes. Importantly, these patterns are being described for the first time in a

documented spatial model of the brain, providing a foundation for future novel inquiry.

In this article, we present the curricular framework, pedagogical considerations, student affective outcomes, and student product assessment from three cohorts of the BM&C course. Where appropriate, we compare outcomes between students enrolled in the BM&C CURE and a non-CURE matched comparison group. Portions of these data have been presented in preliminary form (D'Arcy et al., 2016a,b). A report that focuses on the primary neuroanatomical data generated by multiple cohorts of BM&C students is being prepared separately based on our preliminary reports focusing on the neuroscience aspects of the project (Wells et al., 2015a,b; D'Arcy et al., 2016c; Flores-Robles et al., 2017; Burnett et al., 2018; Martinez et al., 2018; 2019).

1.1: Course requirements

BM&C is an equivalent substitution for the two-semester introductory biology laboratory course sequence. Pre-/co-requisites for the course include the general biology lecture series (BIO1: General Biology and BIO2: Organismal Biology) and a research fundamentals course designed to familiarize students with scientific literature and the inquiry process. It should be noted that, unlike the traditional laboratory courses that meet once per week with no option for extended hours, BM&C meets in two, three-hour sessions per week with additional optional hours offered on an as-needed basis depending upon room and instructor availability.

1.2: Course preparation

In an effort to ensure effective use of class time and to reduce histological error associated with the research process (Simmons & Swanson, 2009), we performed the following pre-course preparations:

a) Adult male Sprague-Dawley rats of consistent body weight at time of sacrifice were used to reduce size variation and to ensure that the sizes of the brains to be studied were similar to that used to produce the Swanson (2004) rat brain atlas.

b) Standardized fixation protocols and chemicals were implemented by A. M.

c) Sections were prepared by A. M. using consistent microtome blade settings (angle and thickness) and a consistent approach to plane-of-section adjustments.

d) The tissue blocking method, tissue section thickness, and the tissue collection scheme were standardized to provide students with well-curated series of experimental tissue.

1.3: Mapping system

As described previously (Khan, 2013; Khan et al., 2018a,b), a major limiting step in the unification of certain kinds of neuroscientific data has been the (largely unintentional) neglect of the neuroscience community to adopt a common spatial framework within which to integrate diverse datasets. This is especially true for datasets in the laboratory rat, a model that has been a mainstay of neuroscience research for at least the better part of a century (*e.g.*, Herrick, 1928; also see Table 4 in Khan, 2013). Indeed, numerous studies have been published on the expression patterns of key macromolecules within the rat brain, but few of these patterns have been mapped

to a standardized brain atlas. This type of mapping for the distributions of macromolecules in the brain allows scientists to contextualize their findings in relation to other datasets mapped to the same reference space, thereby unleashing the predictive potential of chemoarchitectural studies (Khan, 2013; Khan et al., 2018a). Here, we opted to use the Swanson rat brain atlas (2004) as our spatial framework and to train students to perform key experimental and analytical procedures to identify, localize, and map expression patterns of key neuropeptides in the rat brain. We selected the Swanson atlas for several reasons: (1) a few studies, including those conducted by our own multi-institutional collaborative teams, have already been published that have utilized this spatial framework for mapping molecular expression patterns (Swanson et al., 2005; Yao et al., 2005; Geerling & Loewy, 2006; Kerman et al., 2007; Hahn, 2010; Zséli et al., 2016; Santarelli et al., 2018); (2) we have published the first explicitly-documented plane-of-section analysis for activity-dependent gene expression patterns using this atlas (Zséli et al., 2016), an analysis that is taught in this course; and (3) several studies are now taking advantage of the structured nomenclature system for this reference atlas, which can serve as an ontology for use by neuroinformatics specialists and computational neuroscientists seeking to integrate diverse datasets in digital space (see introductory comments in Brown & Swanson, 2013).

1.4: Course outcomes and learning/research objectives

We aligned course objectives to the aforementioned CURE dimensions (collaboration, iteration, scientific practices, discovery, and broader relevance), established CURE-specific outcomes to be assessed (course engagement, science communication, science identity, and science process skills), and identified validated published instruments to be used in evaluating the specified affective outcomes (**Fig. 1, 2**) (Fredickson & Branigan, 2005; Estrada et al., 2011;

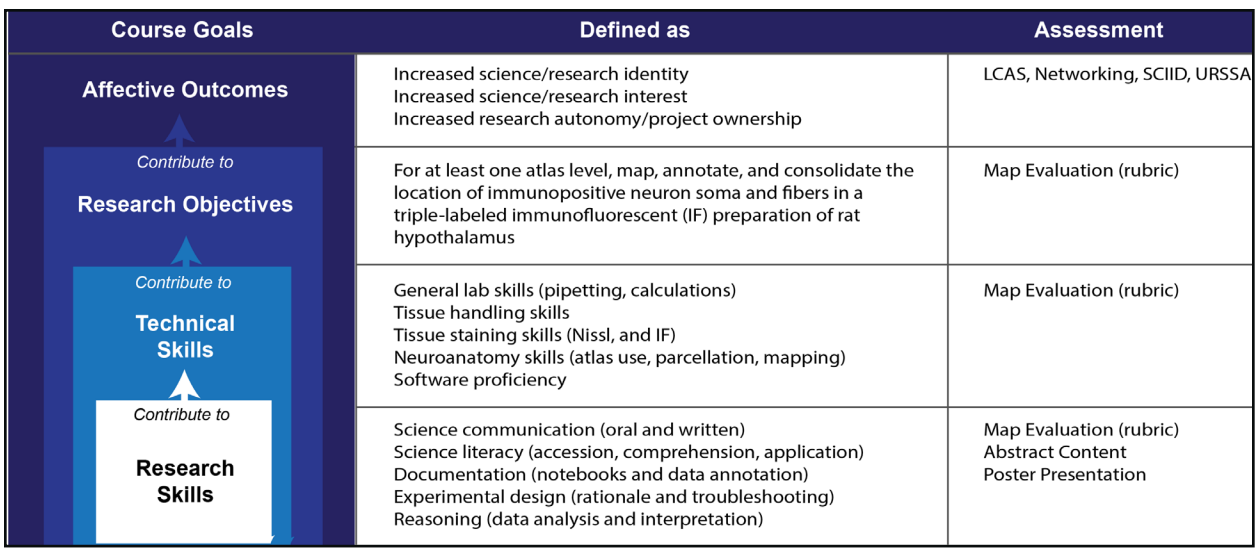

Figure 1. **Course objectives and outcomes**. By enabling students to practice research skills to gain self-sufficiency, to gain proficiency in a specific set of technical skills through iteration, and by setting attainable research goals, the course objectives are designed to yield positive student affective outcomes. Specific benchmarks were identified to describe successful outcomes for each tier of objectives. The mechanism employed to assess each outcome is provided. Laboratory Course Assessment Survey = LCAS; Science Identity Scale = SCIID; Expanded Experimental Design Ability Tool = E-EDAT; Undergraduate Research Student Self-Assessment = URSSA.

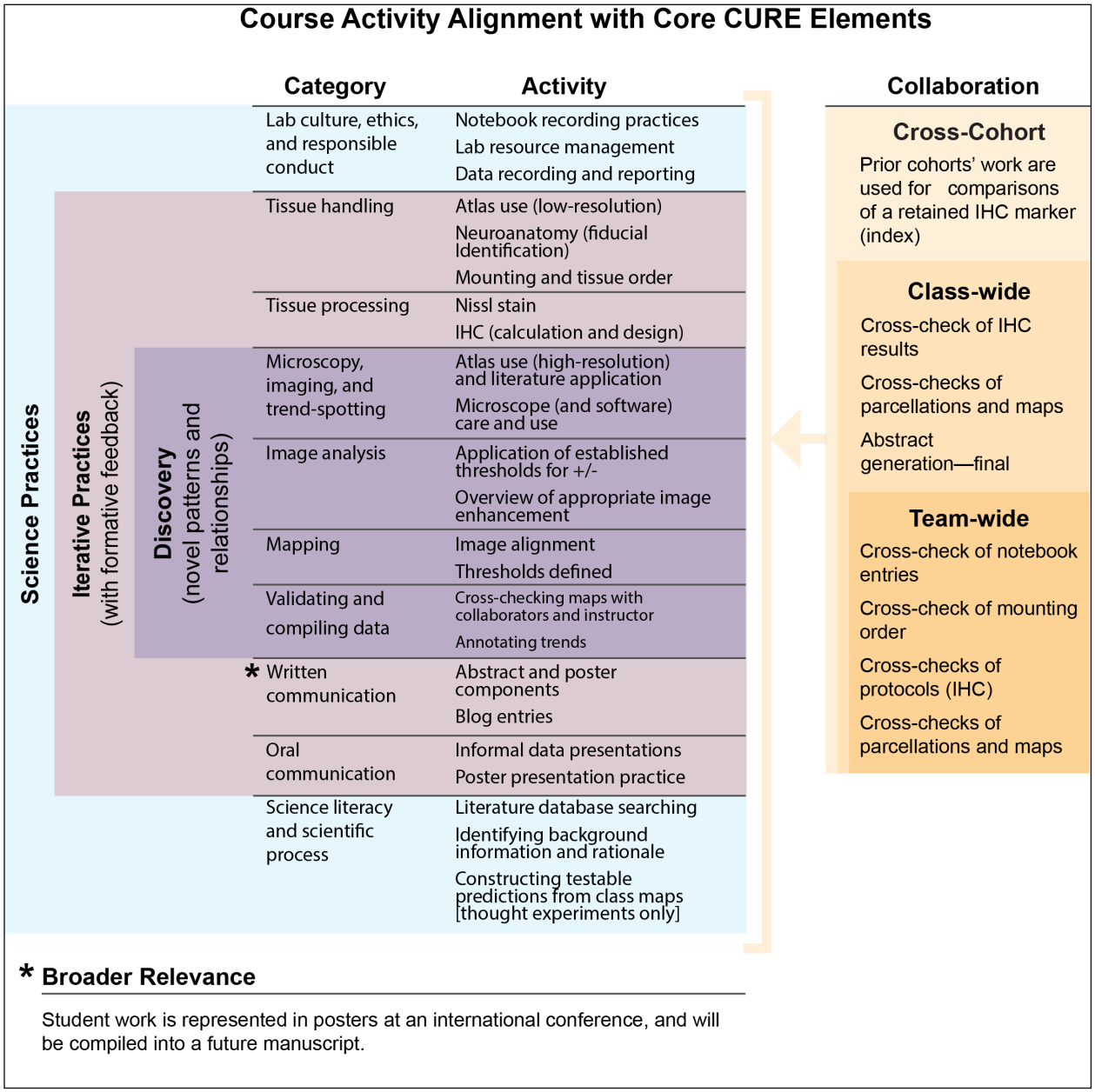

Figure 2. **Alignment of course activities to the five dimensions of CUREs**. Course activities were aligned to the five classic CURE elements: Scientific practices, iterative practices, discovery, collaboration, and broader relevance. Specifically, students iteratively engaged in authentic scientific practices within a tiered framework of collaboration. Their goal was to identify and contextualize novel chemoarchitectural patterns of neurons within hypothalamic regions of interest.

Badalà et al., 2013; Auchincloss et al., 2014; Brownell, et al., 2014; Corwin et al., 2015; Hanauer and Hatfull, 2015; Weston and Laursen, 2015; Rodenbusch et al., 2016). We established clearly-defined, attainable minimum research objectives for the course: 1) to distinguish novel and distinct neuron populations in the rat hypothalamus by their chemoarchitectural properties; and 2) to represent the immunohistochemically-identified cells and fiber distributions in a canonical atlas for at least one complete atlas level. To aid in maintaining consistency across multiple cohorts

taught by four different instructors, we developed a scoring rubric for student-generated maps. We also identified a suite of technical skills specific to the course research project as well as broadly-applicable research skills including documentation, science literacy, and science communication necessary to achieve project success.

1.5: Course description and implementation

1.5.1: Establishing student autonomy. Pedagogy progressively shifted from an instructor-driven style in the first weeks of the course (roughly four sessions in each cohort), to demonstration and application of techniques, and finally to peer training. The peer-training model capitalized upon the staggered progress of individual groups. Teams that completed their protocols early were given instruction in new techniques, which they could then share with other teams, thus providing a means by which focused mentorship could be effectively scaled up while promoting peer teaching and actively demonstrating time-management skills. Finally, students moved toward class-wide collaboration and provision of feedback on generated products (*e.g.*, maps).

1.5.2: Establishing research skills. Students were placed in permanent groups (nmembers = 4) at the beginning of the course and provided with a general introduction to neuroanatomy terminology, the course goals, lab safety, and notebook expectations (**Fig. 2**). Instructors candidly discussed the tissue sample origins and introduced the policies of humane animal handling as well as the IACUC standards for animal care and use. Instructors also informed students that, while care should always be taken in carrying out lab protocols, mistakes inevitably occur. Students were encouraged to treat the errors they made as an important part of their learning process and not something to fear or to obfuscate. In an effort to reinforce this mentality, students participated in developing a strategy for error reporting, recording, and evaluation. Each class session began with a student-driven review of the key points of the preceding lab session, as recorded in their notebooks, and an overview of the current class objectives. Each class session ended with an instructor-led preview of the next set of planned class activities. Instructors used the time during tissue incubation periods to introduce new skills, assign students small projects (thought experiments, abstract and poster development activities, or informal in-class presentations on principles and techniques), and to share interesting news and information relevant to course content.

1.5.3: Immunohistochemistry and tissue processing. Students practiced skills using non-experimental tissue until they reached the desired proficiency level (two rounds practicing the mounting of tissue sections onto microscope slides and two rounds practicing immunohistochemistry provided students with sufficient training and troubleshooting opportunities to proceed on to using experimental tissue). Once student proficiency was sufficient to reduce intra-group variability, instructors assigned an experimental rat brain – pre-cut into 30 μm-thick coronal-plane sections – to each group for further processing. Each team within a cohort performed multi-label immunohistochemistry of rat brain tissue sections for an identical set of three distinct neuropeptide biomarkers **(Table 1)**. This approach afforded students the ability to troubleshoot techniques together as a class, draw comparisons of immunopositive staining across four different animals, and identify robust staining patterns during class-wide examination of preliminary images. Students then worked to image tissue sections in the region bounded by the rostrocaudal extent of the lateral hypothalamic area stitched at ×10 magnification using a Zeiss Axio Imager M.2 epifluorescnce microscope (Carl Zeiss Microscopy, LLC; Thornwood, NY) driven by Neurolucida software

Table 1. Primary antibodies by cohort*

Cohort	Functional association	Target & Host/Type	Antibody registry	Source	Cat #	Dilution
1,3	feeding, arousal, metabolism	melanin-concentrating hormone (MCH); rabbit polyclonal	AB_10013632	Phoenix Pharmaceuticals	H-070-47	1:20K
1,4	responsivity/signalling regulation	NOS1, mouse monoclonal	AB_626757	Santa Cruz	sc-5302	1:5K
1,2,5	feeding, arousal, metabolism	alpha-melanocyte stimulating (aMSH), sheep polyclonal	AB_91683	EMD-Millipore	AB5087	1:10K
2,5	responsivity/signalling regulation	calbindin (CB-955); mouse monoclonal	AB_1140073	Abcam	AB66185	1:1K
2,3,4,5	feeding, arousal, metabolism	orexin A (C-19), goat polyclonal	AB_653610	Santa Cruz	sc-8070	1:4K
3	reward	tyrosine hydroxylase, mouse monoclonal	AB_2201528	EMD-Millipore	MAB318	1:2K
4	feeding, arousal, metabolism	Agouti-related Peptide (AgRP) [82–132]-NH_2 ; rabbit polyclonal	AB_2313908	Phoenix Pharmaceuticals	H-003-53	1:2K

* Students retained at least one common antibody to act as an index for between-cohort variability in staining, imaging, and subject.

(MBF Bioscience; Williston, VT). Students matched immunofluorescence images with images of the adjacent Nissl-stained tissue section for use in parcellation and mapping, as described below.

1.5.4: Neuroanatomical mapping. Instructors introduced Adobe Photoshop CC and Adobe Illustrator CC (Adobe Systems, Inc., San Jose, CA) skills through a series of simple exercises and provided students with a general style template (layer organization and naming, line thickness and marker settings) to scaffold their mapping efforts. Prior to mapping, students were assigned seminal research articles pertaining to the immunohistochemical markers they probed, in relation to the region of interest being examined. This approach was used to not only inform students of the expected outcomes, but also to highlight novel findings as students progressively mapped the regions. This information was then used for the informal, in-class development of novel research questions and experimental design, as well as contextualization of student findings on final posters or oral presentations.

Students were also provided with digital files containing the Swanson (2004) reference atlas and neuroanatomical literature references for the regions of interest in the rat brain. Neuroanatomical boundaries and immunopositive signal (both axonal fibers and neuronal perikarya) were marked directly in Adobe Illustrator CC onto data layers that were superimposed over the layers containing photomicrographic images of immunofluorescence- and Nissl-stained tissue sections (**Fig. 2**). Students then transcribed all data onto the appropriate locations in the Swanson (2004) reference atlas, paying careful attention to preserve the relative distance, distribution, and orientation in the rectified image and performing plane-of-section analysis. Finalized versions of all maps were assessed and validated by a rubric developed for this purpose (**Supplemental Materials II**). As a culminating product for the course, students worked collaboratively with their peers and the course instructor to incorporate mapping data into a conference poster for presentation at the Society for

Neuroscience conference (years 2016–2018).

1.5.5: Assigning grades. Throughout the course, students were assigned the tasks of writing small blog entries, drafting abstract and poster content, and maintaining a laboratory notebook. They were also given occasional concept checks in the form of brief quizzes. Importantly, students were informed that the good-faith effort in producing quality research would be assigned a grade, not the project success. In this way, students were alleviated from the artificial pressure of grades that could potentially tempt them to behave dishonestly (Please refer to the course syllabus: **Supplemental Materials III**).

2. Materials & Methods

2.1: Student population

Students entered BM&C having met the pre-/co-requisite qualifications, as previously described. Three cohorts of BM&C students (n = 42; 79% Hispanic; 79% female) represented majors from multiple STEM disciplines, including: physics, psychology, forensic science, kinesiology, biochemistry, engineering (mechanical, biomedical, electrical), and biological sciences. Twenty-one percent of students were first-generation college attendees, and 50% reported English as their second language. From corresponding non-CURE biology laboratory courses, we drew a matched population for course comparisons, where indicated (n = 42). All participation was strictly voluntary, and surveys were conducted by non-instructor personnel. Students choosing to participate in surveys and data evaluation for the purpose of this study signed an informed consent document granting permission to use their responses in accordance with policies and procedures established by the university's Institutional Review Board (IRB approval #789648). Unless otherwise indicated, all instruments were administered to participants at the start of the semester (pre-semester) and again at the end of the semester (post-semester) for pre-post comparisons.

2.2: Laboratory Course Assessment Survey (LCAS)

To gain a more complete inventory of the course environment, experience, and resources available to students, the LCAS was administered at the end of the course to both BM&C and non-CURE participants (Corwin et al., 2015). Responses to the 17-question survey were provided using a Likert-item scale (1= strong statement disagreement; 5 = strong statement agreement) and questions clustered into three categories (collaboration, discovery, and iteration) for the purposes of analysis. Psychometric data indicated a high level of construct validity and instrument reliability (Cronbach's $\alpha \geq 0.731$ for all categories).

2.3: Networking (NW)

The Networking scale (Hanauer & Hatfull, 2015) was used to evaluate the extent to which BM&C and non-CURE students communicated about course content with individuals outside of the laboratory classroom environment. Student responses were recorded using a five-point Likert-item scale (1= strongly disagree; 5 = strongly agree). Students from both BM&C and non-CURE classes were surveyed at the end of their respective courses.

2.4: Science Identity Scale (SCIID)

We evaluated students' science identity development using the Science Identity Scale employed by Estrada et al. (2011). The SCIID was selected intentionally given its previous use in evaluating historically-minoritized students' science identity development in STEM contexts. Student responses were recorded on a five-point Likert-item scale (1 = strongly disagree; 5 = strongly agree). Surveys were administered to the BM&C and non-CURE cohorts in pre-/post-semester fashion. Psychometric analyses indicated a high level of instrument reliability (Cronbach's $\alpha \geq 0.837$ for both pre- and post-assessment administration of the SCIID).

2.5: Expanded Experimental Design Ability Tool (E-EDAT)

The Expanded Experimental Design Ability Tool (E-EDAT; Brownell et al., 2014) was implemented in pre-/post-semester format to evaluate potential shifts in BM&C and non-CURE students' experimental design skills over the course of the term. Specifically, the E-EDAT presents students with an open-ended prompt that states that an herbal product, ginseng, promotes endurance. Students are then instructed to develop and describe an experiment that tests that claim. Each student response was analyzed by two researchers with expertise in bioeducation and the biological sciences using the published 17-point rubric developed by Brownell and colleagues. High inter-rater reliability was observed between coders ($\kappa = 0.91$; $p < 0.001$), with all disputes resolved by a third researcher with similar expertise.

2.6: Undergraduate Research Student Self-Assessment (URSSA)

At the conclusion of the BM&C course, a modified version of the URSSA (Weston & Laursen, 2015) was administered to gauge students' perceptions of gains in four thematic dimensions related to scientific practices and professional development. These dimensions included: (a) thinking and working like a scientist [science process]; (b) personal gains; (c) research skills; and (d) attitudes and behaviors [engagement]. Student responses were reported on a five-point Likert-item scale (1 = lowest or most negative rating; 5 = highest or most positive ranking). Scaling criteria were dependent upon the dimension being evaluated, as follows: (a) thinking and working like a scientist, personal gains, and skills (no gains - high gains); and (b) attitudes and behaviors (not at all - extremely frequently). Psychometric analyses indicated a high level of construct validity and instrument reliability (Cronbach's $\alpha \geq 0.848$ for all dimensions).

2.7: Statistical analysis

Descriptive statistics were calculated for each dimension presented on the URSSA, with mean item scores reported. LCAS, NW, and SCIID responses were analyzed using a series of Analysis of Covariance (ANCOVA) and Multivariate Analysis of Covariance (MANCOVA) procedures, controlling for student demographics and pre-semester response (where appropriate). E-EDAT analysis for between-group effects was conducted using an ANCOVA, controlling for student demographics and pre-semester E-EDAT score. Paired t-tests were performed to evaluate within-group shifts in E-EDAT performance over the course of the semester. SPSS (v.23) was used to conduct all analyses.

2.8: Neuromapping Scoring Rubric (NSR)

Development of a rubric for the validation of student products was essential to: (1) examine student performance and proficiency with respect to brain mapping; (2) ensure scientific validity of student products; and (3) provide the graduate teaching assistant instructor with a standardized mechanism for scoring student products. Descriptions of each rubric scoring criterion are included in **Supplemental Materials II**. Briefly, rubric scoring fields are designed to account for varying levels of cognitive demand ranging from low-demand tasks such as attention to detail and following instruction (*e.g.*, using assigned formats of layer organization and naming conventions) to high-demand and highly-nuanced tasks (*e.g.*, correctly transcribing immunopositive label from

the location in the experimental brains to the appropriate corresponding location in the atlas). Student-generated maps that represented the hypothalamic region of Level 26 (Swanson, 2004) in its entirety were first de-identified and randomized. A simple 1/0 scoring format was subsequently used to denote the presence or absence of key elements, as detailed in the rubric. Inter-rater reliability was assessed for two scorers ($k = 0.88$; $p < 0.001$), with all disputes resolved through iterative discussion between coders.

3: Results

In order to reduce potential bias associated with variation in laboratory graduate teaching assistant (GTA) instructor for both the BM&C and non-CURE course sections involved in this research, we first analyzed all outcome variables (*e.g.*, LCAS dimensions; science identity) using a Multiple Analysis of Covariance (MANCOVA) procedure. Results indicated no significant difference in student outcomes as a function of laboratory GTA instructor ($F(42, 177) = 1.407$; Wilks' $\Lambda = 0.259$; $p = 0.067$; $\eta p^2 = 0.202$). Furthermore, this effect was found to be true for both those GTAs facilitating the BM&C CURE ($p = 0.573$) as well as those GTAs facilitating non-CURE sections of the laboratory course ($p = 0.199$). Therefore, we aggregated data from all BM&C sections into a single cohort and likewise did the same for all non-CURE sections prior to conducting all subsequent analyses.

3.1: *BM&C students report having greater opportunities to engage in discovery, collaboration, and iteration within the laboratory environment than their non-CURE peers*

CUREs are designed to provide students with an opportunity to engage in novel research that is both collaborative and iterative in nature (Auchincloss et al., 2014). Consequently, we sought to understand the extent to which these design elements were present both within the BM&C CURE as well within the non-CURE learning environment. Students' post-semester Laboratory Course Assessment Survey (LCAS) responses were analyzed using a series of ANCOVA procedures, controlling for student demographic attributes. Results indicated statistically significant, between-group differences on all outcomes: discovery ($F(1, 76) = 13.710$; $p < 0.001$), iteration ($F(1, 78) = 6.340$; $p = 0.014$), and collaboration ($F(1, 72) = 31.680$; $p < 0.001$); with those students enrolled in the BM&C CURE reporting greater opportunities to engage in the aforementioned processes than their non-CURE counterparts (**Fig. 3**).

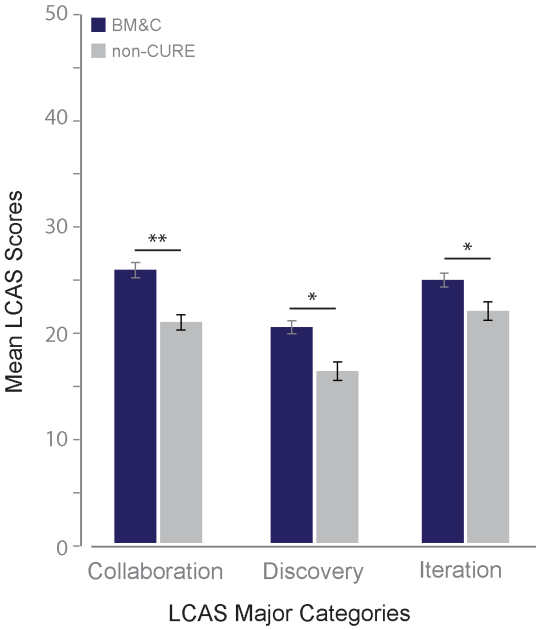

Figure 3. Students' perceptions of the laboratory environment. In a matched comparison of traditional introductory biology laboratory courses (Non-CURE *in gray*) versus the BM&C (*in blue*) courses, BM&C students show significantly higher scores in collaboration (**$p < 0.005$), discovery and iteration categories (*$p < 0.05$).

3.2: *BM&C students communicate about their research to external stakeholders more frequently than their non-CURE peers*

While the LCAS provides a valid measure of within-course collaboration, we were also interested in exploring the degree to which BM&C and non-CURE students communicated information about their research to individuals external to their laboratory course environment. Post-semester data obtained from administration of the Networking Scale (Hanauer & Hatfull, 2015) were analyzed using a series of ANCOVA procedures, controlling for student demographic attributes. Results (**Fig. 4**) showed a statistically significant difference in composite score between

the BM&C and non-CURE cohorts (F(1, 78) = 18.867; $p < 0.001$). Post-hoc MANCOVA analyses further demonstrated that BM&C students discussed their research with parents/guardians (F(1, 78) = 37.350; $p < 0.001$), friends (F(1, 78) = 20.733; $p < 0.001$), other students at their institution not participating in the BM&C course (F(1, 78) = 11.436; $p = 0.001$), and students at other institutions (F(1, 78) = 6.870; $p = 0.011$) to a greater extent than their non-CURE classmates.

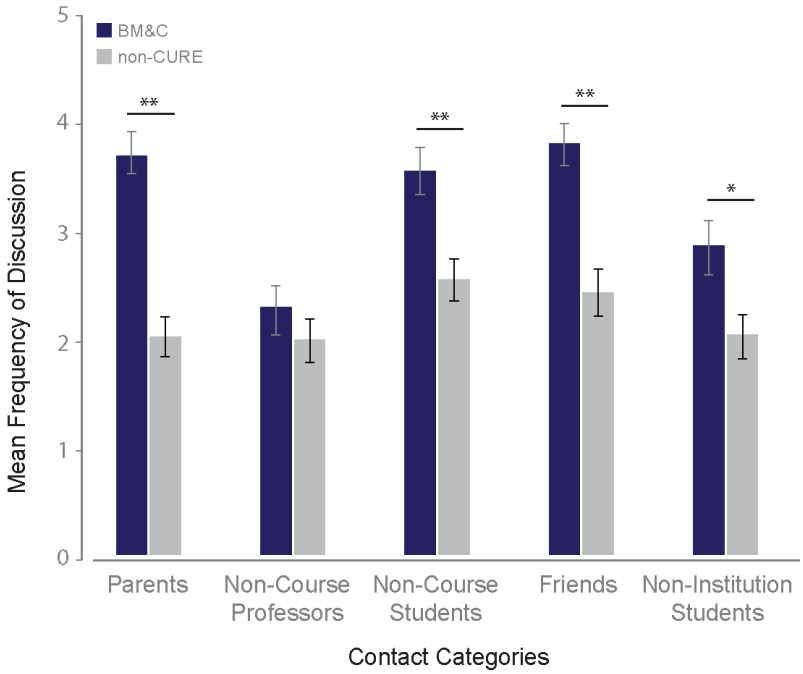

Figure 4. **Student networking patterns**. BM&C students (*blue*) engage in greater levels of external networking than their non-CURE (*gray*) peers (**$p < 0.005$).

3.3: *BM&C students identify as scientists more strongly than their non-CURE counterparts*

Given the role of science identity development as a positive mediator of student engagement, retention, and long-term persistence in STEM (Auchincloss et al., 2014; Estrada et al., 2011), we administered the Science Identity Scale (SCIID; Estrada et al., 2011) to participants in both the BM&C and non-CURE laboratory sections in pre-/post-semester format. ANCOVA analyses, controlling for student demographic attributes and matched pre-semester SCIID response, demonstrated a significant, between-group difference in agreement on a single item: "I have come to think of myself as a scientist" (F(1, 77) = 10.177; $p = 0.002$); with BM&C participants reporting higher average statement agreement (**Fig. 5**).

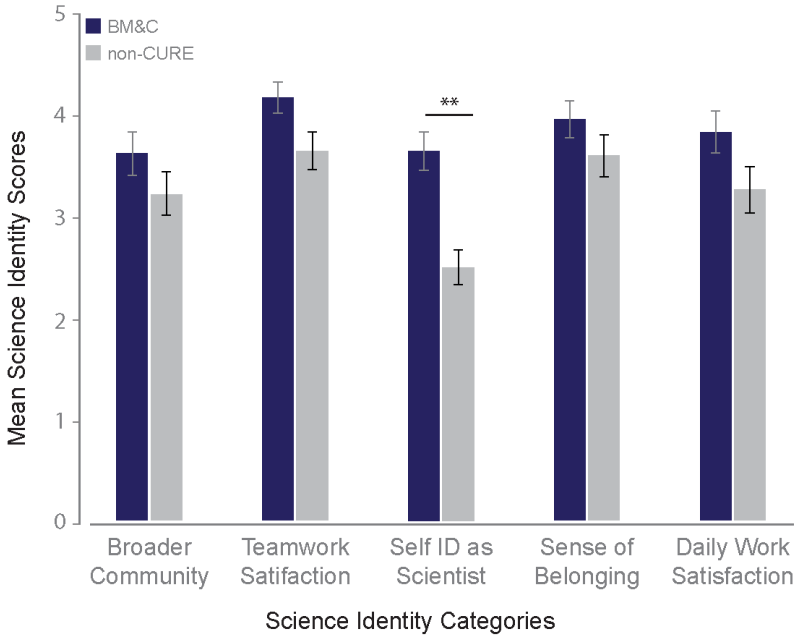

Figure 5. **Science identity scores**. While no differences were seen among many of the categories assessed, BM&C students (*blue*) did self-identify as a scientist more frequently than non-CURE students (*gray*) (**$p < 0.005$).

3.4: *Examining students' development of experimental design competencies*

In an effort to further investigate the impact of the

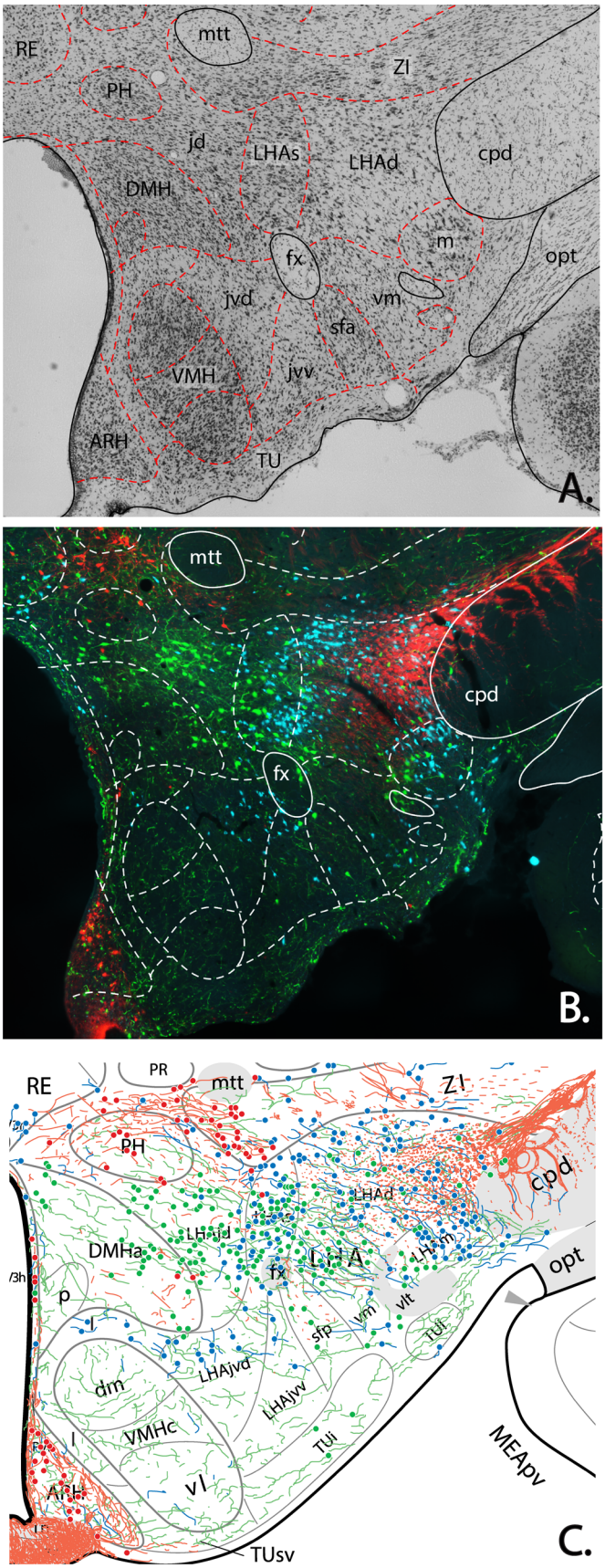

Figure 6. **Sample of student workflow**. **A**. Students identified structures based on the Nissl-stained cytoarchitecture (photographed at ×10 magnification). **B**. Boundaries from the Nissl-stained section were overlaid onto immunohistochemical data images (tyrosine hydroxylase (TH), *red*; melanin concentrating hormone (MCH), *blue*; hypocretin/orexin (H/O), *green*. All images taken at ×10 magnification). **C**. Merged data files of student-generated maps representing fiber density trends and denoting immunoreactive cells within the modeled framework.

BM&C CURE and non-CURE laboratory environments on students' development of experimental design skills, the Expanded Experimental Design Ability Tool (E-EDAT; Brownell et al., 2014) was administered in pre-/post-semester fashion. An ANCOVA analysis, controlling for student demographics and E-EDAT pre-score, revealed a statistically significant, between-group difference in post-score performance ($F(1, 77) = 4.566$; $p = 0.036$) (**Fig. 6**). Interestingly, a posteriori paired t-test analyses indicated no significant shift in pre-post performance for BM&C students ($t(41) = 0.510$; $p = 0.613$); however, a significant decline in performance was observed for the non-CURE cohort ($t(41) = 2.622$; $p = 0.012$).

3.5: *BM&C students demonstrate aptitude in atlas-based neuroanatomical mapping*

Completed maps were obtained from each BM&C student team and evaluated using the Neuromapping Scoring Rubric (NSR) (example rubric evaluation provided in **Supplemental Materials II: Table SM2.1**. Also see **Fig. 6** for student workflow examples), which we developed specifically for this course. Students performed consistently well in the System and Logic category, which evaluated their ability to follow the provided organizational guidelines (layer order, naming, line and marker parameters). Students also generally

exhibited good atlas and anatomical proficiency but required greater feedback and correction on plane of section analysis ($M = 0.75 \pm 0.5$) and boundary identification of certain sub-regions in the hypothalamus ($M = 0.50 \pm 0.6$; *see* **Fig. SM2.3**). Prior to receiving instructor feedback, students likewise experienced difficulty correcting for tissue distortion when translating signal to the final map ($M = 0.50 \pm 0.6$; *see* **Fig. SM2.3**). It is important to note that, due to time constraints, students did not attempt to identify zones of interaction among various molecular expression patterns; as such, no data are available for this rubric criterion.

3.6: *Student perceptions of the BM&C CURE*

Post-semester Undergraduate Research Student Self-Assessment (URSSA; Weston & Laursen, 2015) data were obtained from individuals in the BM&C course in order to provide additional, nuanced detail about students' experiences in the CURE and to provide formative feedback to the BM&C instructional team. Specifically, the URSSA prompts students to self-report gains in four areas: (a) Science Process; (b) Personal Gains; (c) Research Skills; and (d) Engagement. Descriptive analysis of mean scores within the Science Process category indicated that the greatest perceived gains were in finding patterns in data ($M = 4.25$; $SEM = 0.12$) and in being able to relate one's research to his/her coursework ($M = 4.53$; $SEM = 0.10$) **(Fig. 7)**. A similar trend was observed for the Personal Gains-related questions, with BM&C students reporting good gains overall. Specifically, developing patience for the pace of research ($M = 4.44$; $SEM = 0.12$) and understanding what research is actually like ($M = 4.52$; $SEM = 0.11$) received the highest ratings. Subsequent analysis of student responses on Research Skills items demonstrated that BM&C undergraduates reported good gains for the majority of statements. Most notably, this included working with computers, conducting observations, maintaining laboratory notebooks, understanding journal articles, and explaining their project to people outside of their field. With respect to Engagement, participants reported that they frequently engaged in real-world research, felt responsible for their project, felt like a scientist, and thought creatively about their project (**Fig. 7**).

4. Discussion

In this article, we demonstrate that the BM&C course, as implemented, provides students with many of the key positive affective outcomes associated with CUREs, including increased science identity, greater opportunity for advancement in technical skills, and increased research

Theme	URSSA Question	M (SEM)
Science Process	How much did you gain in the following areas as a result of the Brain Connectomics course?	
	Analyzing data for patterns	4.25 (±0.12)
	Figuring out the next step in a research project	4.08 (±0.12)
	Problem-solving in general	4.08 (±0.13)
	Formulating a research question that could be answered with data	3.86 (±0.12)
	Identifying limitations of research methods and designs	4.11 (±0.14)
	Understanding the theory and concepts guiding my research project	4.33 (±0.11)
	Understanding the connections among scientific disciplines	4.13 (±0.15)
	Understanding the relevance of research to my coursework	4.53 (±0.10)
Personal Gains	How much did you gain in the following areas as a result of the Brain Connectomics course?	
	Confidence in my ability to contribute to science	4.19 (±0.12)
	Comfort in discussing scientific concepts with others	4.25 (±0.12)
	Confidence in my ability to do well in future science courses	4.28 (±0.14)
	Ability to work independently	4.28 (±0.12)
	Developing patience with the slow pace of research	4.44 (±0.12)
	Understanding what everyday research is like	4.52 (±0.11)
	Taking greater care in conducting procedures in the lab or field	4.36 (±0.13)
Research Skills	How much did you gain in the following areas as a result of the Brain Connectomics course?	
	Writing scientific reports or papers	3.44 (±0.17)
	Making oral presentations	3.50 (±0.18)
	Defending an argument when asked research-related questions	3.58 (±0.15)
	Explaining my project to people outside the field	4.00 (±0.16)
	Preparing a scientific poster	3.10 (±0.14)
	Keeping a detailed lab notebook	4.00 (±0.14)
	Conducting observations in the lab or field	4.25 (±0.12)
	Using statistics to analyze data	3.30 (±0.22)
	Calibrating instruments needed for measurement	3.53 (±0.21)
	Working with computers	4.25 (±0.13)
	Understanding journal articles	4.11 (±0.13)
	Conducting database or internet searches	3.81 (±0.19)
	Managing my time	3.80 (±0.17)
Engagement	During the Brain Connectomics course how much did you:	
	Engage in real-world science research	4.33 (±0.12)
	Feel like a scientist	4.31 (±0.13)
	Think creatively about the project	4.00 (±0.14)
	Try out new ideas or procedures on your own	3.41 (±0.19)
	Feel responsible for the project	4.33 (±0.13)
	Spend extra hours on research because you were excited about the research	3.70 (±0.21)
	Interact with scientists from outside your school	2.34 (±0.21)
	Feel a part of a scientific community	3.92 (±0.18)

Figure 7. **Post-semester URSSA responses**. Questions are grouped by the theme assessed. M = Mean; SEM = Standard Error of the Mean.

acculturation versus their non-CURE counterparts (Badalà et al., 2013; Hanauer and Hatfull, 2015; Jeffery et al., 2016; Olimpo et al., 2016; Ballen et al., 2017; Frantz et al., 2017). Moreover, these metrics are achieved in a crowd-training environment wherein students can perform high-quality research that can be readily disseminated in traditional circles of professional scientific communication (e.g., scientific conferences; peer-reviewed publications). Indeed, some of the datasets produced by BM&C students have already been showcased at scientific conferences (Wells et al., 2015a, b; D'Arcy et al., 2016c; Burnett et al., 2018; Martinez et al., 2018). Together with the training rubric we have developed for evaluating atlas-based mapping of tissue labeling, this body of work constitutes an initial framework and proof-of-concept that can aid other investigators seeking to develop a full-length teaching laboratory centered upon atlas-based brain mapping at the mesoscopic (tissue-level) scale. Below, we discuss our results in the context of each of the evaluation and assessment mechanisms we employed in this study.

4.1: Impacts of the BM&C course on students' science process skills and science identity development relative to non-CURE participants

The value of the authentic research experience is that it provides students with the ability to repeat a protocol until they achieve a certain level of success or proficiency (iteration), grants students the freedom to pursue novel findings (discovery), and promotes greater interaction with team- and classmates in order to achieve progression in the project (collaboration) (Auchincloss et al., 2014; Corwin et al., 2015). These experiences can influence students' willingness to speak to others outside the classroom about their research or shift the degree to which students perceive themselves to be a member of the scientific research community (Estrada et al., 2011; Hanauer & Hatfull, 2015).

Networking frequency of BM&C students was significantly greater among all peer and non-peer groups, excluding non-course professors, relative to their non-CURE counterparts (**Fig. 4**). This is consistent with Hanauer's and Hatfull's (2015) findings and suggests that BM&C students have personal interest and investment in the course activities to the extent that they commit time outside of class to think about and discuss their research with others. While outside of the scope of the present study, we contend that future research should examine the contextual factors mediating such networking behaviors.

With respect to science identity (Estrada et al., 2011), although BM&C and non-CURE students showed no significant differences in response across the majority of items, scores for both BM&C and non-CURE students were generally positive. Both class formats provided students with a moderate sense of belonging in science and within a broader scientific community. Likewise, students in both cohorts reported agreement with statements regarding teamwork and daily work satisfaction. Importantly, however, BM&C students did report a significantly higher propensity to think of themselves as scientists versus their non-CURE counterparts. Given the often-tedious nature of this form of neuroanatomical mapping, we were encouraged that BM&C students continued to find the daily work of a scientist appealing upon completion of the course.

In addition to examining affective outcomes among BM&C and non-CURE students, we likewise sought to explore the impact of course type on students' development of experimental design skills. Within-group comparisons indicated no significant shift in performance among BM&C students, as measured via the E-EDAT, with pre- and post-semester scores being relatively

equivalent (**Fig. 5**). In contrast, a significant decline in performance was observed for non-CURE participants. Similar outcomes were reported by Goodey and Talgar (2016) in their analysis of the impact of participation in an inquiry-based biochemistry laboratory course on students' experimental design proficiency.

4.2: Trends in student mapping performance

The NSR provided a mechanism by which BM&C instructors could score students' initial mapping attempts and provide students with targeted formative feedback. From a pedagogical standpoint, implementation of the rubric also allowed instructors to identify aspects of the mapping process that posed a challenge for students and, thus, served as areas where future mentoring and feedback were required. For instance, one unanticipated observation was that students could easily identify immunopositive fibers with little need for correction but had intermittent success with cell body (neuronal perikaryon) identification. Often, students would mark cells that were weakly immunopositive (to be denoted with an open circle) as immunopositive (filled circle), thus indicating a need to create a class-wide guide for establishing thresholds. Much of the remaining correction required by students centered on drawing the anatomical boundaries of more subtly-defined sub-regions of the hypothalamus and on applying correction for tissue distortion when transferring data to the final maps. In addressing anatomical boundary issues, instructors used class reference materials to highlight the key features of the cytoarchitecture that defined the regions. In transferring data to digital atlas plates, students required more extensive instruction in adjusting stretch and compression distortions before mastering the ability to correctly translate the spacing, relative positioning, and relative densities of signal. However, students were able to act independently after one or two instructor interventions. Students were also observed providing increasingly meritorious peer critique after receiving instructor input, indicating increased concept mastery and process autonomy.

4.3: Student perception of gains experienced during the BM&C course

Students experienced moderate to good gains in many of the metrics assessed by the URSSA, with highest gains observed in developing patience, conscientious research practices, and understanding the day-to-day realities of authentic research (see Fig. 7). These personal gains have been aligned with student acclimatization to the research environment (Weston & Laursen, 2015). Furthermore, in keeping with the course intent to empower students to engage in authentic research, students of the BM&C course report recognizing their work as "real-world" research, feeling like a scientist, and feeling responsible for their project.

4.4: Scaling up BM&C

Although this study focuses on a small student population, group structures and the use of group members as peer-instructors can provide a means of scaling up class size while maintaining a single course instructor. In order to combine ever-larger sets of data across cohorts, we have developed a tool to assess student products, streamline evaluation and formative feedback, and unify clear expectations for the class. Multiple instructors, who have conducted BM&C courses in parallel to augment crowd-training and expedite data processing, have been aided greatly by

the use of discussion forums and blogs to share feedback and upload systematic evaluation tools, appropriate brain mapping training, and a unified research protocol. Training workshops, in which instructors are equipped with the appropriate starting materials (*e.g.*, tissue sets) along with sample training materials, are recognized as a means to ensure consistency of performance longitudinally as has been done with next-generation sequencing projects (Buonaccorsi et al., 2011, 2014).

4.5: From crowd-trained research to crowd-directed research

The late physicist, Richard Feynman, remarked famously how students can often provide much insight into new research directions, despite having little or no expertise in the subject:

> The questions of the students are often the source of new research. They often ask profound questions that I've thought about at times and then give up on, so to speak, for a while. It wouldn't do me any harm to think about them again and see if I can go any further now. The students may not be able to see the thing that I want to answer, or the subtleties I want to think about, but they remind me of a problem by asking questions in the neighborhood of that problem. It's not easy to remind yourself of these things. (Feynman, (1985), p 166).

From our experiences teaching BM&C, we found numerous occasions where inquisitive students would provide the types of "reminders" Feynman refers to, in which gaps of knowledge within the field are exposed during their discussions with instructors of the course. A promising area of expansion for our course would be to capture and harness the products of such discussions to generate new research questions and to have students help design and conduct new studies based on such questions.

In conclusion, with careful planning and realistic goals, novice undergraduates can generate quality high-spatial resolution chemoarchitectural maps of the brain. Importantly, our findings suggest that a neuroscience focus or major is not an essential requirement for students to derive satisfaction, research identity, and sense of autonomy from conducting this type of research. Providing students with a sense of community, independence, and positive collaboration can influence development of other affective and research-related outcomes.

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D'Arcy *et al*. Supplemental Materials I: Literature Evaluation of CURE and Problem-based Curricula for Neuroscience Undergraduates

In searching extant literature describing neuroscience-themed CURES or inquiry-based approaches, the following search terms were applied in Google Scholar, ERIC, and NCBI's PubMed database search engines: ["Undergraduate" AND "Research" AND "Neuroscience"] or ["Undergraduate" AND "Research" AND "Neuroanatomy"]. Articles were then assessed for content suitability, and courses specifically designed for medical neuroanatomy purposes, neuroscience program descriptions, novel distribution of teaching responsibilities, and non-inquiry-based courses were excluded. **Table SM1.1** (*below*) lists represents a non-exhaustive treatment of the literature, but provides a sense of focus and implementation in readily-available curriculum resources in neuroscience CUREs and problem-based courses.

Table SM1.1: Published reports of curricula and/or implementations of neuroscience CUREs

Reference	Level	Topic/Field	Approach	Implicit Major	Duration	Setting
Bergstrom BP. Using *in vivo* voltammetry to demonstrate drug action: A student laboratory experience in neurochemistry. *J Undergrad Neurosci Educ*, 2012;*10*(2):A113–A117.	UD	Neurochemistry	Fast-scan voltammetry	Yes	Module	Lab
Bodnar RJ, Rotella FM, Loiacono I, *et al*. "C.R.E.A.T.E."-ing unique primary-source research paper assignments for a pleasure and pain course teaching neuroscientific principles in a large general education undergraduate course. *J Undergrad Neurosci Educ*, 2016;*14*(2):A104–A110.	UG	Psychology	Literature/ Writing	No	Course	Class
Brielmaier J. The woman born without a cerebellum: A real-life case adapted for use in an undergraduate developmental and systems neuroscience course. *J Undergrad Neurosci Educ*, 2016;*15*(1):C1–C3.	UG	Systems neuroscience	Case study	Yes	Module	Class

Table notations: UG = undergraduate, UD = upper-division, Novice = freshman-specific, G= graduate, NS = not specified. "Implicit major" = article specifically mentions students enrolled in a specific major as a condition or requisite, Course = a full semester course (partial semesters or multiple semesters indicated for this category). Module = a stand-alone exercise that may span across one to a few class meeting times within a semester-long course.

Table SM1.1 (continued)

Reference	Level	Topic/Field	Approach	Implicit Major	Duration	Setting
Burdo JR. Using chick forebrain neurons to model neurodegeneration and protection in an undergraduate neuroscience laboratory course. *J Undergrad Neurosci Educ*, 2013;*11*(2):A178–A186.	UD	Neuroscience/ Biology	Molecular biology experiments	Yes	Course	Lab
Church WH. Column chromatography analysis of brain tissue: An advanced laboratory exercise for neuroscience majors. *J Undergrad Neurosci Educ*, 2005;*3*(2):A36–A41.	UG	Neurochemistry	Analysis of altered chemistry	Yes	Module	Lab
Crisp KM. A structured-inquiry approach to teaching neurophysiology using computer simulation. *J Undergrad Neurosci Educ*, 2012;*11*(1):A132–A138.	UG	Computational neuroscience	Simulated modeling of directed inquiry	Yes	Module	Class
Dao, V., Yeh, P.-H., Vogel, K. S. and Moore, C. M. Applied neuroanatomy elective to reinforce and promote engagement with neurosensory pathways using interactive and artistic activities. *American Association of Anatomists*, 2015; 8: 166–174.	Novice	Medical neuroanatomy	Interactive activities (drawing)	Yes	Course	Class
Dunne CR, Cillo AR, Glick DR, et al. Structured inquiry-based learning: *Drosophila* GAL4 enhancer trap characterization in an undergraduate laboratory course. Kerfeld CA, ed. *PLoS Biol*. 2014;*12*(12):e1002030.	UD	Molecular cell biology	Gene expression	Yes	Module	Lab
Flint RW, Hill JE, Sandusky LA, Marino CL. Use of the pyrithiamine-induced thiamine deficient animal model of Korsakoff's Syndrome for exploratory research activities in undergraduate physiological psychology. *J Undergrad Neurosci Educ*, 2007;*5*(2):A35–A41.	UG	Neuroscience/ psychology	Animal model behavior	Yes	Course	Lab

Table SM1.1 (continued)

Reference	Level	Topic/Field	Approach	Implicit Major	Duration	Setting
Hall AC, Harrington ME. Experimental methods in neuroscience: An undergraduate neuroscience laboratory course for teaching ethical issues, laboratory techniques, experimental design, and analysis. *J Undergrad Neurosci Educ,* 2003;*2*(1):A1-A7.	UG	Neuroscience	Behavior/ Histology/ Molecular cell experiments	Yes	Course	Lab and class
Kennedy S. Using case studies as a semester-long tool to teach neuroanatomy and structure-function relationships to undergraduates. *J Undergrad Neurosci Educ,* 2013;12(1):A18-A22.	Novice	Neuroscience/ psychology	Case study	No	Course	Class
Khalighinejad B, Long LK and Mesgarani N, "Designing a hands-on brain computer interface laboratory course," *2016 38th Annual International Conference of the IEEE Engineering in Medicine and Biology Society* (EMBC), Orlando, FL, 2016, pp. 3010-3014	Senior/G	Engineering/ biomed	EEG readings	Yes	Course	Lab
Kowalski JR, Hoops GC, Johnson RJ. Implementation of a collaborative series of classroom-based undergraduate research experiences spanning chemical biology, biochemistry, and neurobiology. Hatfull GF, ed. *CBE Life Sci Educ*, 2016;*15*(4):ar55. doi:10.1187/cbe.16-02-0089.	UG	Interdisciplinary	Cross-platform experiments	NS	Course (3 semester)	Lab
Lafer-Sousa R, Conway BR. Vision and art: An interdisciplinary approach to neuroscience education. *J Undergrad Neurosci Educ,* 2009;*8*(1):A10–A17.	UG	Interdisciplinary	Vision, sense, & perception; based data collection	No	Course	Lab and class
Lemons ML. Characterizing mystery cell lines: Student-driven research projects in an undergraduate neuroscience laboratory course. *J Undergrad Neurosci Educ,* 2012;*10*(2):A96–A104.	UD	Neuroscience/ cell biology	Cell line identification	Yes	Course (partial)	Lab

Table SM1.1 (continued)

Reference	Level	Topic/Field	Approach	Implicit Major	Duration	Setting
Lennartz RC. Electrophysiology of the undergraduate neuroscience student: a laboratory exercise in human electromyography. *Amer J Physiol.* 1999; *277*(6Pt2): S42–50.	UG/ novice	Neuroscience/ physiology	EMG readings experiments	Yes	Module	Lab
Lipatova O, Campolattaro MM. The miracle fruit: An undergraduate laboratory exercise in taste sensation and perception. *J Undergrad Neurosci Educ*, 2016;*15*(1):A56–A60.	UG	Sense and perception	Demonstrative experiment	Yes	Module	Lab
Lynd-Balta E. Using literature and innovative assessments to ignite interest and cultivate critical thinking skills in an undergraduate neuroscience course. Allen D, ed. *CBE— Life Sci Educ.* 2006;*5*(2):167–174. doi:10.1187/cbe.05-08-0108.	UD	Interdisciplinary	Literature analysis	Yes	Module	Lab
Meitzen J. Using Tinbergen's four questions as the framework for a neuroscience capstone course. *J Undergrad Neurosci Educ*, 2015;*14*(1):A46–A55.	UD	Neuroscience	Question framework	Yes	Course	Class
Mickley GA, Hoyt DA. Narratives and neurons: Stories of damaged brains. *J Undergrad Neurosci Educ*, 2010;*8*(2):A91–A100.	UG	Neuroscience/ English	Case study analysis	No	Course	Class
Nichols DF. A series of computational neuroscience labs increases comfort with MATLAB. *J Undergrad Neurosci Educ,* 2015;*14*(1):A74–A81.	UG	Psychology/ neuroscience	Software implementation	No	Course	Lab
Nyhus E, Curtis N. Incorporating an ERP project into undergraduate instruction. *J Undergrad Neurosci Educ*, 2016;*14*(2):A91–A96.	UG	Cognitive neuroscience	EEG readings	Yes	Course	Lab
Ogilvie JM, Robbins E. Professor Eric Can't See: A project-based learning case for neurobiology students. *J Undergrad Neurosci Educ*, 2016;*15*(1):C4–C6.	UD	Neuroscience/ psychology	Animal model behavior	Yes	Course	Lab

Table SM1.1 (continued)

Reference	Level	Topic/Field	Approach	Implicit Major	Duration	Setting
Pham K, Romeo RD. An undergraduate laboratory exercise examining the psychomotor stimulant effects of caffeine in laboratory rats. *J Undergrad Neurosci Educ*, 2012;*10*(2):A132–A139.	UG	Neuroscience/ psychology/ behavior/ pharmacology	Behavior/ molecular cell experiments	No	Course	Lab
Raley-Susman KM, Gray JM. Exploration of gerontogenes in the nervous system: A multi-level neurogenomics laboratory module for an intermediate neuroscience and behavior course. *J Undergrad Neurosci Educ*, 2010;*8*(2):A108–A115.	UG	Neuroscience/ behavior	Gene expression/ behavioral outcomes	Yes	Course (partial)	Lab
Roesch LA, Frenzel K. Nora's medulla: A problem-based learning case for neuroscience fundamentals. *J Undergrad Neurosci Educ*, 2016;*14*(2):C1–C3.	UG	Systems neuoscience	Case study	No	Module	Class
Stowasser A, Mohr S, Buschbeck E, Vilinsky I. Electrophysiology meets ecology: Investigating how vision is tuned to the life style of an animal using electroretinography. *J Undergrad Neurosci Educ*, 2015;*13*(3):A234–A243.	UG	Zoology/ neuroscience	ERG readings of field samples	No	Course	Lab
Vilinsky I, Johnson KG. Electroretinograms in *Drosophila*: A robust and genetically accessible electrophysiological system for the undergraduate laboratory. *J Undergrad Neurosci Educ*, 2012;*11*(1):A149–A157.	UG	General neuroscience	ERG reading experiments	Yes	Course	Lab
Willard AM, Brasier DJ. Controversies in neuroscience: A literature-based course for first year undergraduates that improves scientific confidence while teaching concepts. *J Undergrad Neurosci Educ*, 2014;*12*(2):A159–A166.	Novice	Neuroscience, ethics and policy	Literature evaluation	NS	Course (partial)	Class
Yu HJ, Ramos-Goyette S, McCoy JG, Tirrell ME. Community-based, experiential learning for second year neuroscience undergraduates. *J Undergrad Neurosci Educ*, 2013;*12*(1):A53–A60.	Sophomore	Neuroscience (clinical)	Community service and inquiry	Yes	Course	Class/ Community

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Supplemental Materials II:
Scoring and Rubric for Mapping

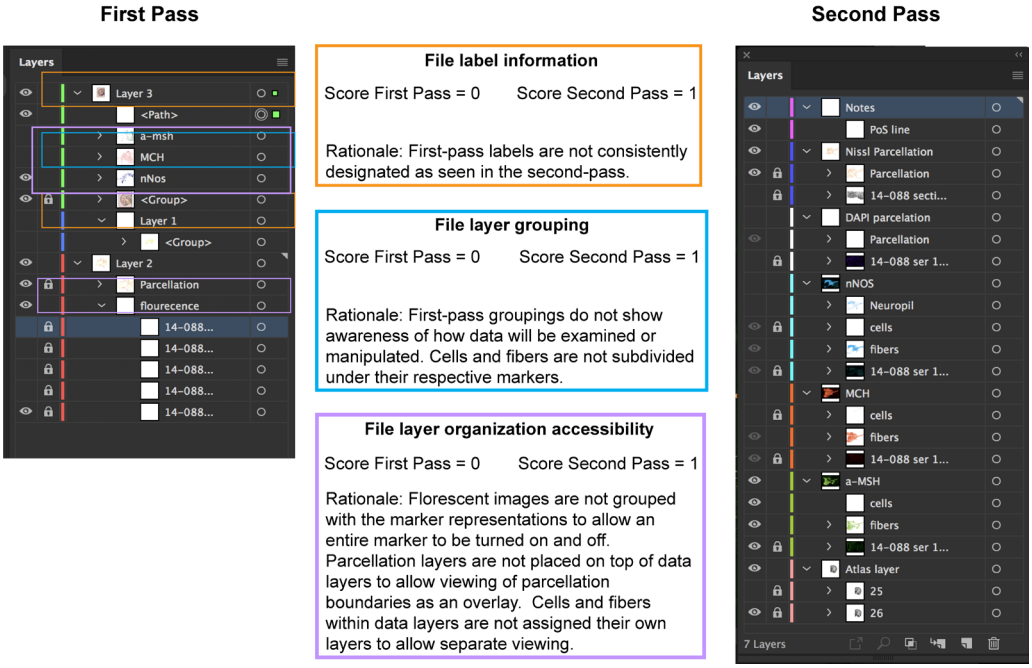

Figure SM2.1: Criteria for scoring, and visual examples of, file management skills taught in Brain Mapping & Connectomics.

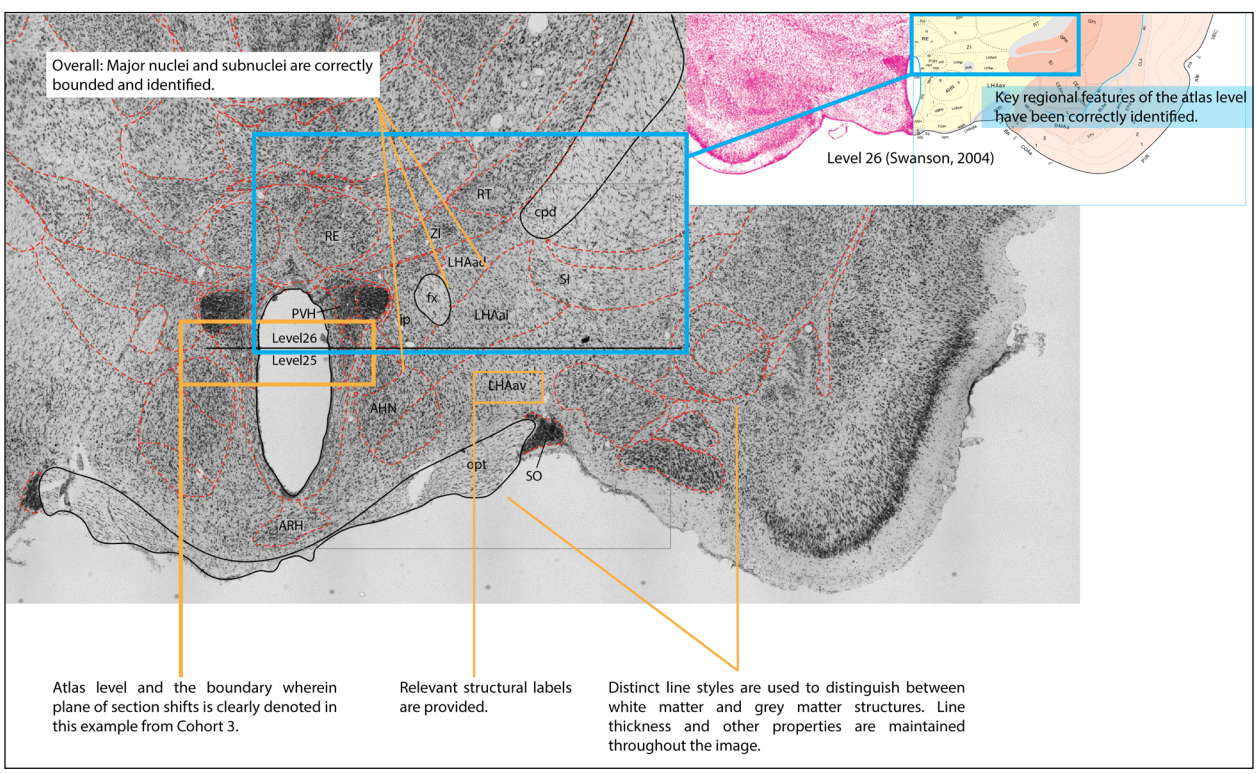

Figure SM2.2: Visual examples of how mapping efforts by students are scored in Brain Mapping & Connectomics.

Table SM2.1. Scoring rubric for atlas-based neuroanatomical mapping

		Group 1	Group 2	Group 3	Group 4	M (SD)
System and Logic	Consistency in demarcation throughout	1	1	1	1	1.00 (±0.0)
	File layers organized accessibly	1	1	1	1	1.00 (±0.0)
	File layers labeled informatively	1	1	1	1	1.00 (±0.0)
	File layer groups organized and labeled logically	1	1	1	1	1.00 (±0.0)
Atlas Proficiency	Dominant atlas level correctly identified	1	1	1	1	1.00 (±0.0)
	Correct atlas level assignment for plane shift	1	1	1[a]	0	0.75 (±0.5)
	Recognition of a shift in plane of section	1	1	1[b]	1	1.00 (±0.0)
	Indicated the axis of change in plane	1	1	1	0	0.75 (±0.5)
Anatomical Proficiency	Major grey matter regions Identified	1	1	1	1	1.00 (±0.0)
	Major grey matter regions correctly bounded	1	1	1	1	1.00 (±0.0)
	Sub-regions identified	1	1	1	0	0.75 (±0.5)
	Sub-regions correctly bounded	0	1	1	0	0.50 (±0.6)
Signal Identification	Complete cataloging of immunopositive cell bodies	1	0	1	1	0.75 (±0.5)
	Complete cataloging of immunopositive fibers	1	1	1	1	1.00 (±0.0)
	Correct signal representation/placement	1	0	1	0	0.50 (±0.6)
	Interaction and localization zones indicated	0	0	0	0	0.00 (±0.0)
	Map Score	**14**	**13**	**15**	**10**	**13 (±2.16)**

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Supplemental Materials III:
Syllabus for Brain Mapping & Connectomics

Brain Mapping and Connectomics Syllabus

Contact information

Scheduled lab hours

Lab goal

The goal of the HHMI Brain Mapping lab is to generate real-world research that is published in a peer-reviewed journal. This makes the Brain Mapping lab unique – you will not just learn a variety of research techniques, and you will not just mimic real research by participating in a semester-long project. You will participate in an actual dissertation project, and your work will be shared with the scientific community. Along the way, you will pick up histological and anatomical research techniques, expand your critical thinking skills, be exposed to professional science writing, and be asked to produce written work in a professional format. In order to achieve the primary course goal of becoming an author on a peer-reviewed scientific publication, you must: be prepared to put in a large amount of time working on the project, and produce high-quality work that meets the stringent standards of the scientific journal this project will be published in.

Project goal

You will be working on the Khan lab's hypothalamic chemoarchitecture project. The goal of this project is to map the distribution patterns of neurons expressing specific chemicals in the rat hypothalamus and to analyze the interactions among these neurons. The hypothalamus is a region of the brain that is famous for participating in control of homeostasis, autonomic and neuroendocrine output, and some motivated behaviors. Disorders of the hypothalamus can potentially affect sleep patterns, metabolic and cardiovascular health, sexual development, and temperament, among other things. There are many different neuronal circuits present in the hypothalamus, but where they are and what form they take is largely unknown, which makes it impossible to understand how the hypothalamus carries out its functions or how it might be disordered in certain medical conditions. Because different neurons express different neurotransmitters, one way to see brain circuits is as a pattern of interacting chemical systems. The hypothalamic chemoarchitecture (chemical architecture) project is concerned with describing, in a very precise and rigorous way, various chemical systems present in the hypothalamus as well as their interactions, as a way of gaining information about hypothalamic circuitry. The project findings will be mapped to a reference atlas (the Swanson 2004 atlas of the adult male rat brain), so that they will be easy for other investigators to use and to integrate with their own data.

Because there are so many different neurotransmitters expressed in the hypothalamus, this is a long-term project which may easily take decades to complete. It will be published as a series of papers that each focus on a particular set of neurotransmitters that we will work together to select as a class.

Student contribution to the project

In groups of four students, you will stain rat brain tissue to visualize functional markers in the brain and map their distribution patterns to the Swanson atlas. Each group will be responsible for staining and mapping one brain. Each group member will be responsible for an equal amount of this work. Some of the mapping will be completed as a group; whatever is left will be divided equally amongst the group members. You will prepare a report of your findings and present your maps to your instructor for quality assessment and inclusion in a paper to be published in a peer-reviewed journal, assuming that quality requirements are met.

Policies

Protection of data: Because you will be conducting actual research, the information you generate in this lab is the intellectual property of [institution omitted]. It does not belong to you. All staining procedures must be written up in your lab notebook, which cannot be removed from the laboratory. You will be allowed to remove the Illustrator files containing your maps from the lab, so that you may work on them at home or at the library, but an up-to-date copy of mapping files must remain on the lab computers at all times. If questions arise amongst the scientific community concerning the staining, your lab notebooks will be referred to. If questions arise as to the integrity of the mapping, your Illustrator files will be referred to. Your lab notebook is considered a legal document. This is serious business.

Required items:

1) Pen
2) Sharpie
3) Number 0 round (or number 0 liner) watercolor brush with natural bristles. The brush tips should not have hairs that splay out to the side and should come to a fine point. Avoid artificial bristles like golden taklon or white nylon. These brushes will be used in manipulating slices of tissue-paper-thin brain, so choose your tool carefully!
4) Lab coat

Excused absences, unexcused absences, and tardiness: Absences may be excused for the following reasons: professional development (such as attendance at a conference), military duties, membership on a [institution omitted] sports team that requires you to leave town for a game, hospitalization or illness, or death in the family. If you know you will need to miss a day of lab, inform your instructor as soon as possible. The instructor must be presented with supporting documentation for any absence to be considered excused, regardless of the reason for the absence. Having an excused absence does not excuse you from completing any assignments that may have been due on that date. Additionally, coordinate with your team members in order to figure out division of

duties, etc., so that your absence does not impose undue burden to your teammates.

One unexcused absence will result in the reduction of your grade by a letter-grade, and two or more unexcused absences will result in an automatic grade of F.

If you are delayed from arriving to class on time and able to safely contact your team members, notify them of what is going on and work with them to resolve any work imbalance. This lab reflects real life as a scientist, and, thus, requires real-life problem solving with your research team.

Academic dishonesty: In accordance with the policies of [institution omitted], academic dishonesty is considered wholly intolerable. Academic dishonesty is essentially any form of cheating. Students found to commit academic dishonesty will be disciplined following [institution omitted] established procedures, which you can read about here: [link omitted].

Plagiarism: Plagiarism is a form of academic dishonesty and will not be tolerated whatsoever. If any part of an assignment submitted by a student is plagiarized, the entire assignment will be considered plagiarized and will count as a zero (0). Particularly, flagrant and deliberate plagiarizers will be referred to the Dean of Students, in accordance with [institution omitted] policies on academic dishonesty.

ADA accessibility: Students with disabilities who require accommodations must contact the Center for Accommodations and Support Services (CASS) as soon as possible at [link omitted]. The CASS office is [location omitted], and their website is [link omitted].

Use of HHMI brain mapping lab equipment: This lab is funded by a grant from the HHMI. Your tuition and lab fees do not contribute to the setup and maintenance of this facility or its equipment. Therefore, your use of the facility and equipment is a privilege, not a right. You may not be present in the lab without the course instructor being present, and you may not use any equipment without the permission of your course instructor. If you violate these stipulations or use the facilities and/or equipment in a disrespectful or irresponsible manner, the TA or course instructor may throw you out of lab and disallow you from attending lab.

Grading

Participation and Attendance	40%
Notebook Check/Quizzes	20%
Project Development and Abstract	20%
Blogging	10%
Final Maps*	10%

Assignments

Participation and attendance: This is a research lab. We are finding the answers to questions no one else has answered before. As such, coming into the lab is essential. Your team, your class, and your collaborating scientist are depending on you to do your part.

Notebook check/quizzes: As a scientist, there is nothing more valuable than keeping clear, detailed, and honest records of what you did and how you did it in order to produce the data you observe, record, and report. Keeping accurate records will be essential, and one of the ways we check notebook content is to conduct notebook-based in-class quizzes. You will have the opportunity to review your notebook contents at the start of class in preparation for the quiz/ potentially use your notebook to furnish information during the quiz.

Blogging: Taking time to think about your science is all a part of the process. Blogging will be similar to your lab notebook in that you will document your progress each week, including any staining that you did and any mapping that you completed. How many sections did you mount? How many sections did you parcellate, etc.? But it is also where you can reflect on problems you encounter, articles you have read, and how your data fits into a bigger picture.

Project development and abstract: Your teams will work together to develop a project to tackle. Part of this will involve reading through scientific papers, and part of it will involve a careful selection of what you will test and how you will set up the experiment (including what areas of the brain we want to examine, why they are important to examine, and what are some signals we want to examine and why). Additionally, we will work together to develop individual, group, and class abstracts that summarize your findings and determine how to present your data on a poster for a professional conference. Participation and demonstration of critical thinking are keys to succeeding in this grade category.

Final maps: For the grade, your instructor will evaluate not the final map, but each step taken to obtain it. Throughout the semester, you will be observed in how well you contribute to your team's maps, your intellectual discussions that take place during the course of mapping, your care in maintaining ordered and clearly-labeled layers, your attention to detail in maintaining line thickness and color, your honesty in noting regions of ambiguity, and your rationale behind your brain region boundaries (parcellations). *It is not about the destination, but the journey.

While not assigned a grade, your final maps will be subject to the critique and examination of the scientific community. Your Illustrator files will be used to assess the quality of your maps by the lab Principle Investigator. High-quality maps will be those that adhere to the reference standards established, that reflect uncertainties in parcellations and transfers carefully, that are assigned to an atlas level(s) correctly and in a thoughtful manner (meaning you didn't just draw a line across the section), and that demonstrate a subtle and refined understanding of the chemoarchitecture underlying parcellations. Such maps will contribute to the poster and future publications. If your maps meet these standards for inclusion, you become eligible for inclusion as an author in subsequent professional communications and reports using this data (see your instructor for further details).

Team Contract

I will be on time.

I will be respectful of my teammates and their time.

I will respect the safety of others and will obey all safety rules and encourage my teammates to do so.

When working as a group, I will contribute intellectually to the task at hand.

I will do my part to complete research outside of lab that my teammates are depending on me for.

I will not allow my teammates to wallow in egregious amounts of my unfinished work.

If I fail in these duties to my team, I understand that my final grade will suffer a loss of points in an amount corresponding to the severity of my failure of my team.

_____

Signature

_____

Printed Name

_____

Date