Synthetic biology offers an ideal opportunity to promote undergraduate laboratory courses with research-style projects, immersing students in an inquiry-based program that enhances the experience of the scientific process. We designed a semester-long, project-based laboratory curriculum using synthetic biology principles to develop a novel sensory device. Students develop subject matter knowledge of molecular genetics and practical skills relevant to molecular biology, recombinant DNA techniques, and information literacy. During the spring semesters of 2014 and 2015, the Synthetic Biology Laboratory Project was delivered to sophomore genetics courses. Using a cloning strategy based on standardized BioBrick genetic “parts,” students construct a “reporter plasmid” expressing a reporter gene (GFP) controlled by a hybrid promoter regulated by the lac-repressor protein (laci). In combination with a “sensor plasmid,” the production of the reporter phenotype is inhibited in the presence of a target environmental agent, arabinose. When arabinose is absent, constitutive GFP expression makes cells glow green. But the presence of arabinose activates a second promoter (pBAD) to produce a lac-repressor protein that will inhibit GFP production. Student learning was assessed relative to five learning objectives, using a student survey administered at the beginning (pre-survey) and end (post-survey) of the course, and an additional 15 open-ended questions from five graded Progress Report assignments collected throughout the course. Students demonstrated significant learning gains (p < 0.05) for all learning outcomes. Ninety percent of students indicated that the Synthetic Biology Laboratory Project enhanced their understanding of molecular genetics. The laboratory project is highly adaptable for both introductory and advanced courses.

INTRODUCTION

The emerging field of synthetic biology provides an ideal opportunity to enhance undergraduate laboratory courses with research-style projects. Synthetic biology is the fusion of multiple scientific fields including molecular biology, systems biology, and engineering (4, 11, 15, 16). Novel genetic systems can be designed and assembled using well-defined genetic elements and standard molecular biology procedures. Professional applications range from industrial products, such as the development of synthetic biofuels (18), to medical treatments and testing, including both pharmaceutical synthesis (22) and diagnostic biomarker detection (9). Using few techniques and inexpensive reagents, undergraduate students can model professional research programs by designing and constructing novel genetic devices in bacteria or yeast. Further, immersing students in an inquiry-based, project-style laboratory course introduces students to the culture of scientific discovery from inspiration to product.

In lieu of single-session, demonstration labs, project-oriented laboratory courses promote better understanding of the scientific process while improving students’ abilities to develop novel research strategies (2, 3, 21, 25, 26). New paradigms in curriculum design advocate for more-active learning modules such as project- and inquiry-based laboratory courses (1, 20). The primary aim of the project involves the students in the design and construction of a simplified “genetic sensor” for the monosaccharide arabinose as a model synthetic biology device. Most exciting however is the enormous potential to fabricate novel synthetic biology devices, and the opportunity for students to establish collaborative, engaging projects.

Intended audience

The molecular biology focus of the project integrates well with a sophomore-level genetics course. The content of the laboratory project has direct implications within
molecular genetics, biotechnology, microbiology, and cellular biology, and can be expanded to upper division courses and undergraduate research opportunities (6, 27).

**Prerequisite student knowledge**

Students should be familiar with the structure and function of DNA and understand the basic principles of gene structure and the processes of DNA replication, transcription and translation. Well-prepared students will recognize that some proteins bind to sequence-specific sites on the DNA to carry out biological functions. Students should have general laboratory knowledge that includes maintaining a laboratory notebook and following basic safety standards.

**Learning time**

The laboratory portion of the genetics course is scheduled for twelve 2.5-hour laboratory sessions (see Table 1). The timing can be compressed if instructors provide intermediate products or skip some lab sessions, or when students have significant molecular biology background.

**Learning objectives (LO)**

Students will develop molecular biology skills through the guided assembly of a genetic device and apply knowledge by independently designing a synthetic biology device. Additional modules require students to evaluate existing genetic devices produced as part of the International Genetically Engineered Machine competition (www.igem.org), and to design a novel sensor system using standardized genetic parts as identified in the Registry of Standard Biological Parts (parts.igem.org).

Upon completion of this laboratory project, students will be able to:

1. Define synthetic biology and its application to address biological problems.
2. Utilize the Registry of Standard Biological Parts and other available databases to find genetic parts of interest and develop a feasible design for a functional device.
3. Apply standard molecular biology techniques to assemble BioBrick parts into “devices” and “systems.”
4. Analyze and evaluate molecular biology data to make conclusions about experimental results.
5. Predict the function of a synthetic biology “device” by reviewing the design.

**PROCEDURE**

The overall goal of the laboratory component of the course is for the students to design and build a genetic system that will 1) provide a readily observable link between genotype and phenotype through a reporter gene, 2) demonstrate genetic interaction of factors in cis- and trans- models, and 3) allow students to explore different facets of synthetic biology through design, construction, and application. The genetic device built by the students includes two plasmids that can be co-transformed into Escherichia coli strain DH5-alpha to act as an environmental sensor and demonstrate the genetic interaction of specific components. The two-plasmid system is composed of a reporter plasmid that will produce a visual phenotypic signal and a sensor plasmid that can respond to environmental arabinose (see Fig. 1).

Students construct a “reporter plasmid” that will express a reporter gene with readily observable phenotype using a promoter regulated by the E. coli lac-repressor protein (lacI gene). A synthetic, hybrid promoter (P_{LacO-I}, part number R0011 in the Registry of Standard Biological Parts) provides a model to discuss the structure-function aspects of prokaryotic promoters. The hybrid promoter is constitutively active but responsive to the lac-repressor protein (12, 19). In order for students to quickly view the phenotype produced by the reporter plasmid, we use the production of fluorescent proteins as the reporter gene. Students are given the choice of green or red fluorescent protein (GFP, RFP), visualized by illuminating cells with a UV-transilluminator or UV-LED flashlight. Colonies producing RFP are frequently tinted red.

Production of the reporter phenotype can be inhibited by a “sensor plasmid” (LU1D7, Table 2) that will produce the lac-repressor protein (lacI) in response to an environmental agent. The specific promoter used in the sensor plasmid will define what the device senses from the environment. We selected the pBAD promoter (14, 23) to construct an environmental sensor keyed to arabinose. Because the pBAD promoter itself is regulated by a trans-acting protein, araC, the sensor plasmid also includes the araC coding region under the control of the constitutively active, endogenous promoter, P_c (14). Each plasmid must be maintained within the cells using separate selective markers (8), therefore the reporter plasmid is maintained through an ampicillin resistance marker (amp^R) while the sensor plasmid utilizes the kanamycin resistance marker (kan^R).

When combined in the same bacterial cell, the two plasmids produce a single genetic “device” that will produce differential responses in the presence or absence of arabinose (Fig. 1). In the absence of arabinose, transcription from the pBAD promoter is limited to basal levels. Without lac-repressor activity, the P_{LacO-I} will activate, expressing the reporter gene (GFP or RFP). In the presence of arabinose, an arabinose-araC protein complex will activate transcription from the pBAD promoter to produce the lac-repressor protein and inhibit transcription from the P_{LacO-I} promoter, reducing fluorescence. Thus, green or red fluorescent cells indicate the absence of arabinose, while in the presence of arabinose, cells will not fluoresce.
TABLE 1. Synthetic biology lab schedule.

<table>
<thead>
<tr>
<th>Week</th>
<th>Scheduled Lab</th>
<th>Concept Development</th>
<th>Student Activity</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Laboratory Safety, Lab 1: Introduction to Synthetic Biology</td>
<td>Compare previous synthetic biology projects.</td>
<td>Online research of iGEM database.</td>
</tr>
<tr>
<td>3</td>
<td>Lab 3: PCR Color Genes –“Red” or “Green”</td>
<td>PCR reaction mechanism and components, primer design.</td>
<td>Introduce micropipetting skills. Prepare PCR reactions, thermal cycler.</td>
</tr>
<tr>
<td>4</td>
<td>Lab 4: PCR: Gel Electrophoresis and DNA Purification, Registry Website</td>
<td>Gel electrophoresis theory and practice, DNA purification.</td>
<td>Agarose gel for PCR reactions. Purify PCR-generated DNA with spin-columns.</td>
</tr>
<tr>
<td>5</td>
<td>Lab 5: BioBrick Assembly: Restriction Enzyme (RE) Digestion</td>
<td>Review BioBrick system for plasmid assembly. Simulate methods with models.</td>
<td>Digest PCR generated and purified DNA to produce the reporter plasmid “Insert.”</td>
</tr>
<tr>
<td>7</td>
<td>Lab 7: Reporter Plasmid: Plasmid DNA Extraction and Screening</td>
<td>Discuss need to screen transformed cells for ideal product.</td>
<td>Prepare plasmid DNA from transformed cells to identify reporter plasmid.</td>
</tr>
<tr>
<td>8</td>
<td>Lab 8: DNA Sequencing: Reporter Plasmid</td>
<td>Discuss DNA sequencing methods and data analysis.</td>
<td>Use sample or actual data for DNA sequence analysis using online tools.</td>
</tr>
<tr>
<td>9</td>
<td>Lab 9: Sensor Plasmid: Novel Plasmid Design</td>
<td>Review breadth of synthetic biology ideas present in iGEM database (online).</td>
<td>Use examples from the iGEM database to design a novel sensor plasmid (online).</td>
</tr>
<tr>
<td>10</td>
<td>Lab 9 (cont’d): Sensor Plasmid: Student Design Presentations</td>
<td></td>
<td>Students present and defend their sensor plasmid design.</td>
</tr>
<tr>
<td>12</td>
<td>Lab 10 (cont’d): Device Analysis: Compare Gene Expression Outcomes</td>
<td></td>
<td>Score the activity of the sensor device in the presence/absence of arabinose.</td>
</tr>
</tbody>
</table>

PCR = polymerase chain reaction; iGEM = International Genetically Engineered Machine.

The Synthetic Biology Laboratory Project is divided into 12 weekly sessions for the topics shown. The Synthetic Biology Laboratory Manual includes modules developing each weekly topic (“Scheduled Lab” column, Appendix I). Weekly topics are supported by “Concept Development” discussions and exercises and “Student Activity” elements including wet-lab or skills practice sessions. The table does not address gaps for institutional breaks or other student assignments outside of the laboratory project.

Materials

We adopted the BioBrick system (7, 16, 17) to simplify the recombinant DNA methodology. The system utilizes a standardized structure of genetic elements to support rapid plasmid assembly and provide a consistent format to introduce molecular biology techniques in an undergraduate environment. Each of the BioBrick parts is an independent genetic element, such as promoters, ribosome binding sites, coding regions, and transcriptional terminators. Each part is flanked by a BioBrick prefix (5’ end of the part) and suffix (3’ end of the part) that contains restriction enzyme recognition sites used for plasmid assembly and mapping reactions. The structure of the BioBrick part, including the prefix and suffix, is diagrammed in the Synthetic Biology Laboratory Manual (Appendix I, Fig. 2.3). BioBrick part assembly uses a standardized set of reactions and restriction enzyme sites (review Appendix I, Module 5). By adopting a standardized “part” structure and methodology, students are able to focus on the design of the genetic project instead of repeatedly finding new enzyme sites for the next assembly reaction.

Techniques require standard molecular biology enzymes and equipment (Taq DNA polymerase, micropipettors and tips, thermal cycler, horizontal gel boxes, agarose, UV-transilluminator, etc.) as well as bacterial growth (media preparation, incubation, and disposal). Three commercial kits are recommended for the required enzymes and DNA ligase (NEB BioBrick Assembly Kit, NEB-E0546), DNA fragment purification (IBI Scientific Gel/PCR DNA Fragment Extraction Kit, IB470300), and plasmid extraction from E. coli cells (IBI Scientific Hi-Speed Mini Plasmid Kit, IB47101). Optionally, DNA sequencing of student products can be completed by commercial sources. Computers with Internet access are needed to obtain information from online...
resources and analysis tools; no proprietary software is required. Online iGEM databases provide ample background information on the BioBrick system and synthetic biology, as well as an immense variety of projects designed and implemented by undergraduate students.

A collection of genetic elements (BioBricks) used in the course project is presented in Table 2. To expand the project or develop novel BioBrick materials, individuals can obtain more materials from the Registry of Standardized Biological Parts as a public, subscription-based, resource (igem.org/Labs_Program). Readers may contact the authors to obtain a minimal collection of BioBrick materials required to present the course.

Student instructions

Detailed instructions are included in the Synthetic Biology Laboratory Manual (Appendix 1) for students to complete the steps to construct a novel plasmid and analyze their products (Fig. 2). Sequential laboratory modules define the stepwise construction and verification of the reporter plasmid and sensor device (Table 1).

Faculty instructions

Twelve weekly sessions include concept development, practical skills, and wet-lab activities (Table 1). Lab sections of 24 students were divided into 12 student pairs to work together. For each 2.5-hour lab session, detailed faculty instructions for each of the 10 lab modules including materials and specific preparation information are included in the Instructor’s Manual and Materials List (Appendix 2). Lab modules can be divided into three conceptual sections: introduction to the system and basic techniques (weeks 1–3), guided construction and verification of a reporter plasmid (weeks 4–8), and design and integration of a sensor plasmid to complete the device (weeks 9–12).

To become familiar with synthetic biology, students explore previous iGEM competition projects during initial lab sessions, and then present examples to the class. During weeks 3 to 7, conceptual material focuses on the theory and practice of recombinant DNA techniques before application in wet labs to assemble the reporter plasmid. In week 8 students complete a “walkthrough” of DNA sequence analysis using model data (Appendix 3) or optional sequencing results. Finally in weeks 9 to 12, students design an environmental sensor using BioBrick parts from the Registry of Standard Biological Parts. The students present their design and evaluate the designs presented by their peers (week 10). Due to time constraints, students do not construct their designed plasmids, but will co-transform their reporter plasmid with a prepared plasmid containing the arabinose sensor (LU1D7, Table 2). Transformation reactions including the sensor and reporter plasmid plated onto media with and without arabinose will be used to determine the final
activity of the sensor device (weeks 11–12). Optionally, novel BioBrick parts created by the students can be submitted to the Registry of Standard Biological Parts as a student “publication.”

Suggestions for determining student learning

Assessment of student learning relative to the five Learning Objectives is accomplished through an anonymous survey of the students given at the beginning (pre-survey) and completion (post-survey) of the course (Appendix 4). The student survey includes 16 questions relating to content knowledge (Q 1–12), student perceptions of synthetic biology (Q 13, 14), and student opinions about the curriculum (Q 15, 16). The 16 questions assess subject matter knowledge of synthetic biology and the BioBrick system (Q 1–6), technical skills relevant to molecular biology techniques (Q 7, 8, 11, 12), information literacy (Q 9, 10), the interdisciplinary nature of synthetic biology (Q 13), social implications of synthetic biology (Q 14), and preferential impact of the course on the students (Q 15, 16).

Additionally, five graded Progress Report assignments include free-response questions that specifically address experimental and procedural outcomes as well as assessment of learning outcomes. The series of five Progress Report assignments are included in Appendix 5, and are keyed to be completed in weeks 3, 5, 7, 10, and 12. Student responses to select questions in these assignments are mapped to the five Learning Outcomes and can be evaluated on a point-based rubric reflecting core concepts (Appendix 6).

### TABLE 2.
Genetic elements required for the student project.

<table>
<thead>
<tr>
<th>Local ID</th>
<th>Part Description</th>
<th>Registry ID</th>
<th>iGEM Distribution</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Reporter Plasmid: Assembled in pSB1A3</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LU1A8</td>
<td>R0011 Synthetic Promoter: lacI regulated, lambda pL hybrid</td>
<td>BBa_R0011</td>
<td>2010 1 6G</td>
</tr>
<tr>
<td>LU1B3</td>
<td>Ribosome Binding Site: RBS</td>
<td>BBa_B0034</td>
<td>2010 1 2M</td>
</tr>
<tr>
<td>LU1A6</td>
<td>Green Fluorescent Protein: GFP</td>
<td>BBa_K145015</td>
<td>2010 2 2L</td>
</tr>
<tr>
<td>LU1B1</td>
<td>Monomeric Red Fluorescent Protein: mRFP</td>
<td>BBa_E1010</td>
<td>2010 1 18F</td>
</tr>
<tr>
<td>LU1B8</td>
<td>Composite Intermediate: B0034 + K145015</td>
<td>Longwood construct</td>
<td>— — —</td>
</tr>
<tr>
<td>LU1C4</td>
<td>Composite Intermediate: B0034 + E1010</td>
<td>Longwood construct</td>
<td>— — —</td>
</tr>
<tr>
<td>LU1C8</td>
<td>Reporter Plasmid (GFP) R0011 + LU1B8</td>
<td>Longwood construct</td>
<td>— — —</td>
</tr>
<tr>
<td>LU1E4</td>
<td>Reporter Plasmid (RFP) R0011 + LU1C4</td>
<td>Longwood construct</td>
<td>— — —</td>
</tr>
<tr>
<td><strong>Sensor Plasmid: Assembled in pSB1K3</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LU1A4</td>
<td>Promoter: pBAD</td>
<td>BBa_I13453</td>
<td>2010 1 1F</td>
</tr>
<tr>
<td>LU1A5</td>
<td>Lac-Repressor Protein: lacI</td>
<td>BBa_C0012</td>
<td>2010 1 2O</td>
</tr>
<tr>
<td>LU1C7</td>
<td>araC Expression Cassette (promoter with operon)</td>
<td>BBa_I13458</td>
<td>2010 1 13L</td>
</tr>
<tr>
<td>LU1C3</td>
<td>Composite Intermediate: B0034 + C0012</td>
<td>Longwood construct</td>
<td>— — —</td>
</tr>
<tr>
<td>LU1D7</td>
<td>Sensor Plasmid (Arabinose) I13458+I13453+B0034+C0012</td>
<td>Longwood construct</td>
<td>— — —</td>
</tr>
<tr>
<td><strong>Plasmid Vectors:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LU1B6</td>
<td>pSB1A3 (AMP-resistant cloning vector)</td>
<td>pSB1A3</td>
<td>2011 — —</td>
</tr>
<tr>
<td>LU1B7</td>
<td>pSB1K3 (KAN-resistant cloning vector)</td>
<td>pSB1K3</td>
<td>2011 — —</td>
</tr>
</tbody>
</table>

The parts used to construct the reporter plasmid and the sensor plasmid for the student project are listed. “Registry ID” indicates the identification number for parts listed in the Registry of Standard Parts (http://parts.igem.org). Materials stored locally are numbered as shown in “Local ID.” The “Part Description” contains base names for genetic elements or a list of items assembled into composite parts. More detailed descriptions, sequence information, and devices using each part are available online by searching the Registry of Standard Biological Parts using the Registry ID. The specific distribution information for the DNA material used in the project is listed in the column labeled “iGEM Distribution.” Plasmid maps diagramming the reporter and sensor plasmids are included in Figure 1.
Various homework assignments can be used to develop content and assess student learning. Brief, in-class examples with answer keys are included in the student and instructor's manuals (Appendices 1 and 2). Literature review assignments can be included early in the course to review published synthetic biology–based research projects or later in the course to investigate the specific genetic elements and interactions within the device.

Sample data

The ungraded reports of five students are included (Appendix 7) for each of the Progress Report assignments as examples of student work. A complete set of sample DNA sequencing results from the first iteration of the course is included for review and student analysis in Appendix 3.

Safety issues

Safety issues are primarily related to general lab safety, chemical hygiene, and biosafety considerations. For all laboratory-based exercises, general laboratory safety practices include the use of safety glasses or goggles, wearing closed-toed shoes, hand-washing at the beginning and end of the lab, prohibition of food and drinks, and the restriction of dangling items such as jewelry or long hair. Significant hazards include working with live *E. coli* bacteria (13), the mutagen ethidium bromide, and exposure to UV illumination. Institutional chemical disposal requirements should be confirmed before beginning each lab. Note that SYBR Green is a safer alternative to ethidium bromide for visualizing DNA in agarose gels (5). For labs requiring live bacteria, the American Society for Microbiology has published biohazard safety guidelines for undergraduate laboratories (13) which are also available at the following URL (http://www.asm.org/index.php/education2/22-education/8308-new-version-available-for-comment-guidelines-for-best-biosafety-practices-in-teaching-laboratories).

In addition to the general practices above, to minimize contamination, students should wear lab coats and not handle personal items. Lab spaces should have nonporous bench and floor surfaces and be sterilized before and after lab sessions. Detailed safety information is included for each laboratory module (Appendices 1 and 2).

DISCUSSION

Field testing

The synthetic biology laboratory course was delivered to genetics courses in the spring semesters of 2014 and 2015. The course often includes multiple sections (three in 2014 and two in 2015) and is team-taught with all course sections participating in the Synthetic Biology Laboratory Project.
Students in the field test include a mixture of sophomores (2014: 38%, 2015: 53%), juniors (2014: 48%, 2015: 35%), and seniors (2014: 14%, 2015: 12%). Use of human subjects for assessment of the Synthetic Biology Laboratory Project was approved by the Longwood University Institutional Review Board for both offerings.

Evidence of student learning

For the 2014 genetics course, 27 of an initial 30 students completed the course and answered the post-survey; for 2015, 32 of an initial 35 students completed the course. Survey results for the two semesters are combined and presented as the percent correct for each subject matter question (Fig. 3). A single-tailed t-test was performed to compare the pre and post scores for each item. All items except question 2 demonstrated significant increases in student learning gains (p < 0.05) following their completion of the laboratory curriculum.

The student survey administered in 2014 and 2015 is prone to errors resulting from students guessing at answers or providing random responses on the survey. For the 2015 semester, we identified questions from the Progress Reports to provide an additional instrument for the course assessment. Materials from 2014 were not retained and could not be reviewed. Fifteen items from the Progress Reports were identified that correlated with Learning Objectives 1 to 5 (Table 3). The Progress Reports were the primary evaluation of students’ ability to evaluate molecular biology data (LO 4). For each criterion, learning outcomes were considered “met” if the average score (reported as the 95% confidence interval for the sample average) was equal to or greater than 70% of the maximum possible score (Table 3). Using this analysis, 13 of the 15 criteria scored “met,” with four criteria significantly exceeding the 70% threshold (Z-test; p < 0.05).

Performance toward different objectives

The results of the assessment program demonstrate that students gain knowledge and develop skills relative to Learning Objectives 1 through 5, with significant increases in abilities for most objectives. One major goal for this novel curriculum is to promote the ideals of synthetic biology while introducing molecular biology at an introductory level. The course focuses on teaching students to interpret synthetic biology designs in terms of genetic interactions, to predict function from design, and to apply their knowledge to developing their own devices. These goals are embodied within the five Learning Objectives (LO 1–5). Survey question 11 integrates the ability of students to recognize genetic symbols (as in questions 3–6) and to interpret the genetic function in terms of a model gene. Correct student responses increased 54% (p < 0.001), demonstrating significant learning gains. Similarly, activities in lab modules 9 and 10 to develop a new device are assessed in Progress Report #5, questions 3–5. Evaluation of student designs of the sensor plasmid (Q 4 or 5) and a new device (Q 3) demonstrate student competence in correctly attributing function to genetic elements and the construction of functional gene models, and designing novel, coherent, and functional genetic devices (Table 3).

The student survey assessed perceptions about the interdisciplinary aspect of synthetic biology (Q 13), the potential application of synthetic biology (Q 14), and student opinions about the lab project (Q 15, 16). Because the genetics course has a mixture of students from different academic years, which may affect their preconceptions, we surveyed first-year students from a prerequisite course at the end of the 2014 and 2015 semesters (Appendix 8). Among all student groups, more than 80% of the students completing survey question 13 indicated that Cell Biology and Genetics/Molecular Biology have a strong impact on synthetic biology. Similarly, fewer than 50% of all students considered that less biologically oriented fields such as physics, mathematics, and computer science have a strong impact. In contrast, students who had completed the synthetic biology curriculum demonstrated a marked increase in their expectation that the Engineering (22%) and Graphic Design/Technical Writing (8%) fields provide a strong impact on synthetic biology (Appendix 8).
Similarly, students completing the Synthetic Biology Laboratory Project foresaw a greater range of applications for synthetic biology in all of the potential applications listed in the survey. More than 60% of all student groups agreed with the potential applications of synthetic biology listed in the survey, and this percentage increased to 94% for those students completing the course (Appendix 8). Topics such as “Production of new energy sources” and “Development of new medicines” were directly addressed during class discussions, so strong agreement for these items was expected. However, “Agricultural practices” and “Detoxify hazardous material from the environment” were not well represented in the course, yet also demonstrated increased acceptance by the students. Thus the current course reinforces the idea that synthetic biology–based devices can be used to address many socially important issues.

Student evaluation regarding the structure of the lab course (Q 15) indicated that 56% of students felt that the synthetic biology laboratory “contributed to their understanding of molecular biology and genetics,” and 50% of students preferred the project-based laboratory (Q 16) to the unit-based (n = 62). Anecdotal comments from students unhappy with the project-based methodology reported that they “got lost when the lab went on for too long.” These results are similar to other assessments of project-based labs (10, 21, 24) and may reflect the “growing pains” of changing formats. In contrast, other students were eager to continue using synthetic biology as a model for other course projects or requesting more experience by seeking out additional research opportunities.

Our results indicate that the contribution of traditionally non-biology fields like mathematics and computer science to the modeling and engineering aspect of synthetic biology as well as other “big data” domains of modern biological practices is not being well developed in the introductory courses. However, after participating in

### TABLE 3.
Assessment of Content Knowledge using Progress Report Assignments.

<table>
<thead>
<tr>
<th>Mapped LO</th>
<th>Progress Report</th>
<th>Question</th>
<th>Average Score</th>
<th>70% Target Score</th>
<th>Possible</th>
<th>Outcome</th>
<th>n</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1.61 ± 0.22</td>
<td>1.4</td>
<td>2</td>
<td>met*</td>
<td>33</td>
</tr>
<tr>
<td>2</td>
<td>1</td>
<td>3a–c</td>
<td>2.52 ± 0.19</td>
<td>2.1</td>
<td>3</td>
<td>met†</td>
<td>33</td>
</tr>
<tr>
<td>3</td>
<td>1</td>
<td>4</td>
<td>2.21 ± 0.3</td>
<td>2.1</td>
<td>3</td>
<td>met</td>
<td>33</td>
</tr>
<tr>
<td>2</td>
<td>2</td>
<td>1a, c</td>
<td>2.13 ± 0.34</td>
<td>2.1</td>
<td>3</td>
<td>met</td>
<td>30</td>
</tr>
<tr>
<td>3</td>
<td>2</td>
<td>1d</td>
<td>2.37 ± 0.45</td>
<td>2.8</td>
<td>4</td>
<td>met</td>
<td>30</td>
</tr>
<tr>
<td>5</td>
<td>3</td>
<td>3a,b</td>
<td>3.39 ± 0.24</td>
<td>2.8</td>
<td>4</td>
<td>met†</td>
<td>23</td>
</tr>
<tr>
<td>4</td>
<td>3</td>
<td>3c,e</td>
<td>1.83 ± 0.21</td>
<td>2.1</td>
<td>3</td>
<td>not met</td>
<td>30</td>
</tr>
<tr>
<td>4</td>
<td>4</td>
<td>1c</td>
<td>2.03 ± 0.32</td>
<td>2.1</td>
<td>3</td>
<td>met</td>
<td>31</td>
</tr>
<tr>
<td>4</td>
<td>4</td>
<td>2c</td>
<td>1.97 ± 0.36</td>
<td>2.1</td>
<td>3</td>
<td>met</td>
<td>31</td>
</tr>
<tr>
<td>5</td>
<td>5</td>
<td>1a</td>
<td>3.24 ± 0.38</td>
<td>2.8</td>
<td>4</td>
<td>met*</td>
<td>33</td>
</tr>
<tr>
<td>3</td>
<td>5</td>
<td>1b</td>
<td>2.12 ± 0.32</td>
<td>2.1</td>
<td>3</td>
<td>met</td>
<td>33</td>
</tr>
<tr>
<td>2</td>
<td>5</td>
<td>1c</td>
<td>1.29 ± 0.31</td>
<td>1.4</td>
<td>2</td>
<td>met</td>
<td>21</td>
</tr>
<tr>
<td>3</td>
<td>5</td>
<td>1d</td>
<td>1.39 ± 0.19</td>
<td>2.1</td>
<td>3</td>
<td>not met</td>
<td>33</td>
</tr>
<tr>
<td>5</td>
<td>5</td>
<td>4</td>
<td>2.76 ± 0.44</td>
<td>2.8</td>
<td>4</td>
<td>met</td>
<td>33</td>
</tr>
<tr>
<td>5</td>
<td>5</td>
<td>3,4,5</td>
<td>2.52 ± 0.43</td>
<td>2.8</td>
<td>4</td>
<td>met</td>
<td>33</td>
</tr>
</tbody>
</table>

LO = learning outcomes.
The learning outcomes are mapped to 15 specific open-ended questions assigned via the Progress Reports (Appendix 5) and evaluated using content-based rubrics (Appendix 6). Student performance is listed as an average score with a 95% confidence interval and compared to the 70% target score of the maximum possible score to determine whether each criterion is “met” or “not met.”

1Learning outcomes, see text for numbered list.
2Specific questions from Progress Reports (Appendix 5).
395% confidence interval for the average values shown.
4n = the number of responses evaluated for each question. Student scores significantly greater than the 70% target are indicated as *: p < 0.05; †: p < 0.001.
the laboratory program, students demonstrate a greater understanding of the interdisciplinary nature of science and can perceive a greater application for synthetic biology to address real-world issues. Scientific literacy of the American public requires an educated population that is not only able to accurately interpret scientific information but to independently reach conclusions consistent with the data and prevailing models. We believe that by fostering an understanding of synthetic biology and molecular biology techniques in current and future generations of students, they will be better able to cope with upcoming changes in medical science, environmental trends, and political policy.

Possible modifications

The laboratory itself is highly flexible to account for student experience and background as well as course limitations such as time or learning objectives. The Synthetic Biology Laboratory course has strong topical roots in molecular genetics and can facilitate learning goals in bacterial genetics, prokaryotic gene structure, and molecular biology; it is therefore amenable to a variety of different courses. To shorten the schedule, some lab modules may be omitted or converted to a homework assignment. For instance, Lab Module 8 on DNA Sequence Analysis could be skipped, converted into a lecture example, or reassigned into a homework assignment. Some early lab modules are included as introductory or informational and may not be necessary for more advanced students. Other labs can be completed by the instructor or skipped if the instructor elects to provide students with intermediate products.

The broad array of genetic elements in the Registry of Standard Biological Parts provides an astounding potential to create novel devices. While the current project challenges students to design and construct a pre-planned project, the actual device produced can be modified by the instructor or student teams. As included in the first lab module, reviewing projects from previous iGEM competitions will inspire both instructors and students to design, build, and test novel synthetic biology devices. The generalized methods presented in the Synthetic Biology Laboratory Manual are applicable to the assembly of any plasmid from BioBrick parts. As presented in Lab Module 10, simple sensors and detectors can be constructed from the many different promoter/operator systems available for student projects, and they might sense metals, sugars, or other suitable agents. For more advanced courses, the project can be modified to build complex systems such as digital logic gates (AND, OR, NOR, XOR, etc.) or more intricate sensory circuits such as a stable, bipolar switch mimicking the bacteriophage lambda regulatory system. Also, students and interdisciplinary faculty from biology, chemistry, computer science, and mathematics can team up to design and build a novel synthetic biology device to compete in the annual iGEM Giant Jamboree.

SUPPLEMENTAL MATERIALS

Appendix 1: Synthetic biology laboratory manual
Appendix 2: Instructor’s manual and materials list
Appendix 3: Sample DNA sequencing data
Appendix 4: Synthetic biology survey with key
Appendix 5: Progress report assignments and keys
Appendix 6: Progress report assessment rubrics
Appendix 7: Examples of student work
Appendix 8: Student perceptions survey response

ACKNOWLEDGMENTS

The authors would like to thank Dr. Malcolm Campbell of Davidson University for his mentorship in synthetic biology and the Genome Consortium for Active Teaching (GCAT) program for providing the workshops that introduced us to synthetic biology. We wish to thank the New England Biolabs Educational Course Support program for the generous gift of reagents and materials. We also appreciate the work of Dr. Andy Anderson and Jennifer Beach in critically reviewing the manuscript. We wish to thank our students for taking part in this exercise, especially those allowing us to present their work: Katy Bell, Andrew Cressman, Kyle George, Hailey Kintz, and Kalyn Steigerwald. The authors declare that there are no conflicts of interest.

REFERENCES


