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ARTICLE

Demonstrating core molecular biology principles using GST-GFP in a semester-long laboratory course

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Abstract

Undergraduate laboratory courses are essential to teaching core principles in STEM. This course, Quantitative Biological Methods, provides a unique approach to teaching molecular biology research techniques to students, in a laboratory that is delivered in a sequence that parallels standard biomedical research laboratory protocols. Students attend a lecture where they are taught the essential principles of biomedical research, and a lab where they learn to use laboratory equipment, perform experiments, and purify and quantify DNA and proteins. The course begins with an introduction to laboratory safety, pipetting, centrifugation, spectrophotometry, and other basic laboratory techniques. Next, the lab focuses on the purification and analysis of glutathione S-transferase (GST) fused to green fluorescent protein (GFP) from an *Escherichia coli* lysate. Students study this GST-GFP fusion protein and perform protein quantification, enzyme assays, chromatography, fluorescent detection, normalization, SDS-PAGE, and western blotting. Students then learn recombinant DNA technology using the GST-GFP vector that was the source of the fusion protein in the prior labs, and perform ligation, transformation of *E. coli* cells, blue/white screening, DNA purification via a miniprep, PCR, DNA quantification, restriction enzyme digestion, and agarose gel electrophoresis. Students write laboratory reports to demonstrate an understanding of the principles of the laboratory methods, and they must present and critically analyze their data. The lab methods described herein aim to emphasize the core molecular biology principles and techniques, prepare students for work in a biomedical research laboratory, and introduce students to both GST and GFP, two versatile laboratory proteins.

KEYWORDS

molecular biology laboratory course, protein purification and analysis, recombinant DNA technology, undergraduate curriculum

1 | INTRODUCTION

Molecular biology research is a highly specialized field which requires years of coursework and laboratory training to master. Aspiring researchers can benefit from learning both theoretical knowledge and practical skills in biomedical research. In this 11-week course, students are introduced to many essential laboratory techniques that are presented through a stepwise sequence of protocols meant to simulate a research environment, as they learn to quantify, purify, and analyze DNA and proteins. The lab provides hands-on laboratory experience utilizing established, broadly applicable research protocols, while additionally taking a novel approach to teaching students about glutathione S-transferase (GST) and green fluorescent protein (GFP), which are frequently used in both molecular biology research and teaching laboratories.

1.1 | Importance of teaching a molecular biology laboratory

Undergraduate STEM courses are a fundamental step in the education of future scientists, in a growing pool of interdisciplinary fields. Various sets of laboratory skills may be field-specific or selectively utilized, and the incorporation of a wide array of foundational research techniques (such as recombinant DNA technology and protein quantification) into a single course provide undergraduate students with the opportunity to learn a broad set of research skills.

This course, Quantitative Biological Methods, provides students with the opportunity to follow a directed path through molecular biology techniques, presented to emulate the realistic nature of biomedical research by incorporating sequential laboratory protocols. Other laboratory courses have successfully implemented a continuous lab format, such as utilizing the various features of GFP to create a set of linked laboratory experiments for students to follow.[1–4] The experimental protocols proposed in this paper expand on this idea by combining two research-relevant proteins, the enzyme GST and a fluorescent protein GFP, allowing students to study both together as a GST-GFP fusion protein in numerous assays.

This setup creates a laboratory environment with a diverse selection of available protocols, many of which fit within the recommendations for a biochemistry lab by the American Chemical Society.[5] In course-based undergraduate research experiences (CUREs), analysis of primary data by students results in enhanced learning benefits compared to those who are given sample data.[6] While not a CURE, during this lab, students analyze their results and make decisions based on their data, which can vary between groups. Students also complete lab reports and quizzes to demonstrate a deeper understanding of the protocols and principles.

Some methods have been optimized to fit within the limits of a four-hour laboratory section, such as the use of a TCE stain in SDS-PAGE, which allows students to visualize proteins immediately and subsequently perform western blotting without the need for a second gel.[7] Many steps can be further modified to adapt the lab to the time and equipment available at other institutions, while maintaining the core principles of the course. Protocol modifications are typically first optimized through a CURE called PILOT.[8]

1.2 | Relevance to research

In this lab, students learn various DNA- and protein-based techniques to quantify, purify, and analyze a GST-GFP fusion vector and protein. GST is involved in the detoxification of xenobiotics,[9] and plays an important role in cellular pathways (such as jun kinase inhibition) and in certain pathologies (such as asthma and cutaneous basal cell carcinoma).[10] In the research lab, GST is often tagged to proteins for affinity purification or enzymatic tracking. It has also been established as a useful undergraduate laboratory teaching tool, due to various aspects of the protein's structure, such as its small size, stability, solubility, enzymatic activity, and binding specificity which allows for affinity chromatography.[11]

Green fluorescent protein, naturally expressed in *Aequorea* jellyfish, was discovered in 1961 by Nobel Laureate Osamu Shimomura et al.[12,13] In 1979, the structure of its chromophore was determined, and in 1992 GFP was cloned and recombinantly synthesized in *Escherichia coli*.[12] Many GFP variants are now available to scientists, which aid in biomedical research (Such as its incorporation into a fluorescent calcium sensor).[13–15] GFP is used in transgenic animals, reporter genes, plants, microbial research, and many other applications.[14] Due to the extensive information available on GFP and the inherent benefits of its fluorescent properties for detection, it is already in use in many published teaching lab protocols, and various experiments have been designed using GFP, including PCR, agarose gel electrophoresis, bacterial transformation, chromatography, and SDS-PAGE.[1–4]

Combining GST and GFP allows students to study both of these important proteins in the same course. GST-GFP can be analyzed and quantified using typical laboratory techniques (protein quantification, chromatography, SDS-PAGE, western blotting); additionally, GST can be studied using affinity chromatography and a 1-chloro-2,4-dinitrobenzene (CDNB) enzymatic assay, and GFP utilizing its chromogenic nature.[1,11] Both can also be analyzed in comparison to controls using anti-GST and anti-GFP antibodies. Additionally, the vector

containing the GST-GFP gene can be used to study recombinant DNA techniques. This diverse range of protocols allows students to use GST-GFP to become familiar with many essential molecular biology and biochemistry lab techniques.

The laboratory curriculum introduces students to many important research techniques, beginning with micropipetting, centrifugation, and spectrophotometry, as well as data collection and analysis. Students are given an *E. coli* lysate containing a 6xHis-GST-TEV-EGFP fusion protein, which allows for various methods of quantification, purification, and analysis. In the protein section of the lab course, students perform protein quantification, enzyme assays, affinity chromatography, ion exchange chromatography, fluorescence detection, sample normalization, generate a purification table, perform protein crystallization, SDS-PAGE, western blotting, and buffer preparation. Next, in the DNA section of the course, students ligate a GST-GFP fragment into a vector, perform a transformation, blue/white screening, a miniprep, DNA quantification, PCR, restriction enzyme digestion, and agarose gel electrophoresis. Students gain hands-on experience with many essential molecular biology techniques, with the added benefit of learning about GST and GFP, two proteins which they may encounter in a research laboratory.

2 | MATERIALS AND METHODS

2.1 | Laboratory overview

This core laboratory course, Quantitative Biological Methods, is taught to juniors at the University of Central Florida (Orlando, FL). Students are required to have completed Biology I and Chemistry Fundamentals I and II. While the lecture enrollment can reach over 450 students, the laboratory sections are limited to ~25 students each, providing students a hands-on lab experience. Students attend three 50-min traditional lectures (which explores the deeper principles behind the techniques) and one 4-h laboratory section weekly.

Prior to each lab, students are expected to review the previous lab's material and read the current lab's protocol. Students take a pre-lab quiz either prior to lab or in the first 10 min. Next, the instructor gives a presentation on the principles of the methods, an overview of the protocol, laboratory safety, and important instructions on the proper usage of new laboratory equipment. Students partner with the person sitting next to them and work in teams of two with the same partner the entire semester. Together the group performs the protocols using the materials provided, while the instructor and teaching assistants answer questions and manage equipment. Students analyze their data together in the lab with their partner and the instructors to ensure a thorough understanding of the results, and then they each submit an independent lab report the following week. After each lab, students complete a post-lab quiz.

2.2 | Protocol overview

An overview of the laboratory protocol is show in Figure 1. Because most undergraduate students have no prior molecular biology laboratory experience, we find that this order (analyzing the protein, followed by the DNA) offers several advantages. The content can be delivered more slowly, as each protein technique takes an entire lab section, whereas each DNA lab often covers three or more techniques. Additionally, the protein-based techniques offer more practice pipetting various volumes early in the semester, with more room for error, while the DNA labs often have only a few precise pipetting steps. Finally, some of the protein-based techniques (such as SDS-PAGE) take longer to understand, perform, and analyze, and having these earlier in the semester is beneficial. However, for advanced students, these labs could be completed with the DNA section prior to the protein section.

In the first section (Lab 1), students are introduced to basic laboratory techniques, including laboratory safety, micropipetting, mixing and vortexing, centrifugation, spectrophotometry, and data analysis.

In the protein section (Labs 2–8), students learn to purify, quantify, and analyze the GST-GFP fusion protein from an *E. coli* lysate. They begin by performing protein quantification via Lowry and Bradford Assays, which allows students to compare and contrast the two techniques. Next students perform a CDNB enzyme assay to establish a GST's reaction velocity in the sample for future comparisons. Students then purify GST-GFP via affinity and ion exchange chromatography, and identify which fractions contain the protein of interest via the chromatogram, a CDNB enzyme assay, and fluorescence. Next the samples are quantified via a plate reader, analyzed using a purification table, and prepared for SDS-PAGE. Students also perform protein crystallization. Finally, GST-GFP is analyzed via SDS-PAGE and western blotting.

In the DNA section (Labs 9–11), students learn essential recombinant DNA techniques, beginning by ligating a GST-GFP fragment into a vector and performing an *E. coli* transformation. The recombinant bacteria are analyzed via blue/white screening and the vector is purified via miniprep. Students quantify this DNA and perform PCR. Finally, students perform restriction enzyme

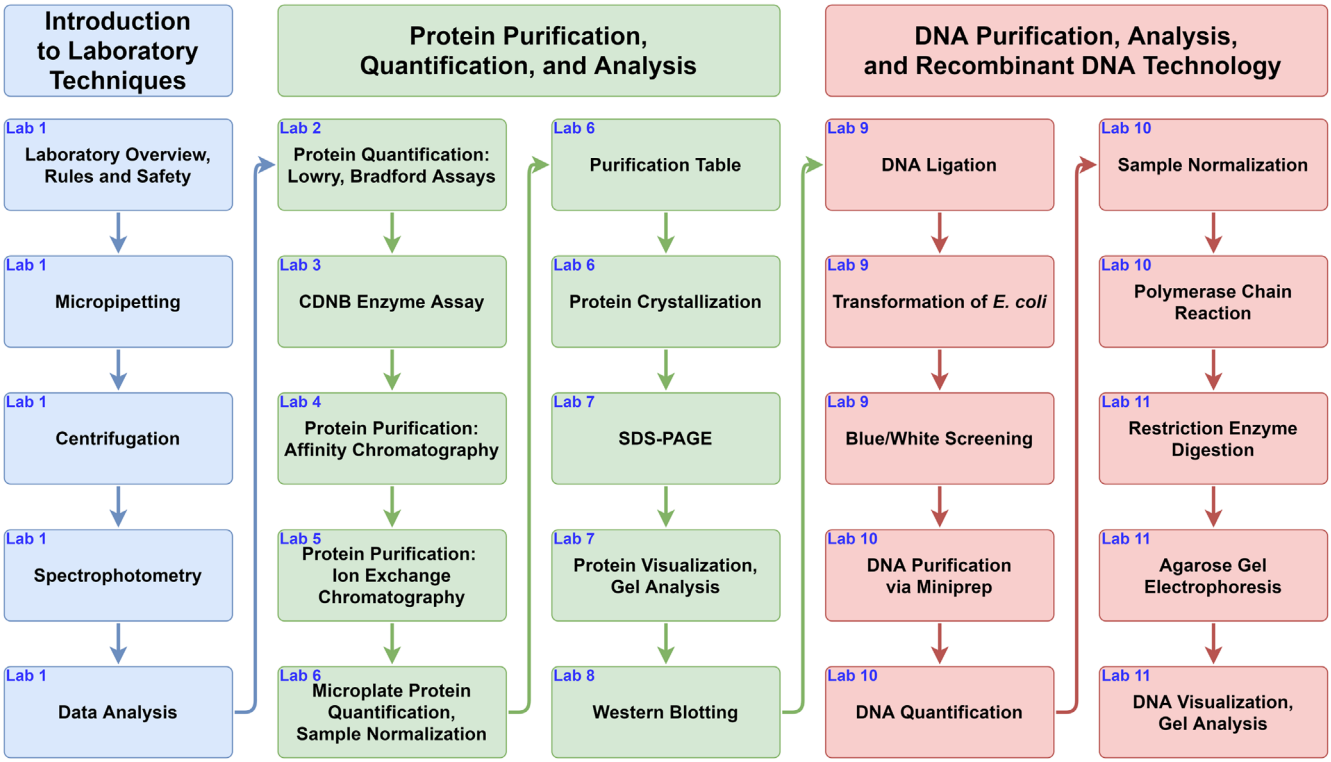

FIGURE 1 Laboratory overview. The laboratory is divided into three sections by color, with the lab number and techniques listed. In section 1 (blue), students are taught lab safety, the basics of lab equipment, and how to analyze their data. In section 2 (green), students purify, quantify, and analyze the GST-GFP protein through quantification, chromatography, SDS-PAGE, and western blotting. In section 3 (red), students use recombinant DNA technology to ligate GST-GFP DNA into a vector, transform bacteria, purify the DNA, and analyze the results with PCR and agarose gel electrophoresis. Flowchart made with Draw.io

digestion of the vector and analyze the results using agarose gel electrophoresis.

2.3 | Lab design and objectives

Students participate in 11 weekly laboratory sessions, each of which lasts approximately 3–4 h. A complete list of materials, equipment, and faculty preparation are included in Supplementary Data 1, and student protocols are included in Supplementary Data 2. Faculty can prepare an introductory lecture for each lab to introduce the principles of the techniques and to provide an overview of the protocol, and a lab quiz can be given on current and prior material depending on the course design.

2.3.1 | Section 1: Introduction to laboratory techniques

Lab 1: Micropipetting, centrifugation, and spectrophotometry

In Lab 1, students are introduced to laboratory safety and basic techniques. Students learn how to use Denville and Eppendorf micropipettes, including applying a tip, setting a defined volume, and aspirating and dispensing liquids using the first and second stop. They are also taught proper technique when handling and labeling microcentrifuge tubes.

Next, students perform a pipetting protocol to practice pipetting various amounts of different liquids. Samples are then vortexed, centrifuged, and transferred into cuvettes. Absorbances are measured in a spectrophotometer at 595 nm, which corresponds to the amount of a blue solution added to each sample. Students plot absorbances in Excel, add a line of best fit, calculate the R^2, and analyze their pipetting consistency in a lab report.

2.3.2 | Section 2: Protein purification, quantification, and analysis

Lab 2: Protein quantification: Lowry and Bradford assays

In Lab 2, students learn protein quantification via the Lowry and Bradford Assays. Students are provided with ~1 mg/mL lysed *E. coli* cells which contain an overexpressed protein induced from a pET expression vector containing a 6×

histidine tag, glutathione S-transferase, a TEV cleavage site, and enhanced green fluorescent protein (GST-GFP).

In the Lowry Assay, students make several BSA standards to generate reference absorbances for a standard curve. The Lowry Assay is performed on both the BSA standards and *E. coli* lysate dilutions, and absorbances are recorded at 750 nm. The BSA standard absorbances are plotted and used to calculate the *E. coli* lysate concentration. The Bradford Assay demonstrates an alternative quantification method without the use of a standard curve (which could be added if preferred). *E. coli* lysate dilutions are pipetted into Bradford Reagent and absorbances are recorded at 595 nm, and absorbances are used to directly estimate protein concentration. Students then analyze and compare the results from both assays.

Lab 3: CDNB enzyme assay

In Lab 3, the *E. coli* lysate is analyzed for GST enzymatic activity by calculating its reaction velocity (units/ml). Students prepare solutions containing two GST substrates: 1-chloro-2,4-dinitrobenzene (CDNB), and glutathione (GSH). Upon adding several dilutions of the lysate to various cuvettes, students record an increase of absorbance at 340 nm over time. GST activity, or reaction velocity, is determined for each dilution, and a final average reaction velocity calculated (typically ~3 units/mL). Students also measure and plot the decreased reaction rate as the reaction approaches equilibrium. GST activity will be used in subsequent labs to locate the enzyme during purification, and to determine the specific activity and fold purification.

Lab 4: Protein purification: affinity chromatography

In Lab 4, students purify GST-GFP from the *E. coli* lysate via affinity chromatography. The lysate is loaded onto a GST affinity column (a His column could also be used) and attached to a chromatography machine. A gradient of GSH is used to elute GST-GFP into fractions, which is located using the A_{280} from the chromatogram. Students also test fractions for GST-GFP via the CDNB Assay to determine reaction velocities, and additionally by using a Gel Doc to detect GFP fluorescence. The A_{280} will indicate the location of all protein, the enzyme assay only GST, and the Gel Doc imaging only GFP. Therefore, taken together, all three methodologies provide unique information for the students to analyze and utilize when determining which fractions to keep. Students pool fractions that contain GST-GFP for further purification and analysis.

Lab 5: Protein purification: ion exchange chromatography

In Lab 5, students perform ion exchange chromatography (IEX) to further purify GST-GFP from the affinity fractions kept from the previous lab. Students load these pooled fractions onto an anion exchange column, and a gradient of NaCl is used to elute GST-GFP into fractions. The chromatography machine measures each fraction's A_{280} to detect protein, and conductivity to measure the salt concentration of the buffer. The fractions are further analyzed with both the CDNB Assay and the Gel Doc, to determine the location of both GST and GFP. Students keep the fraction that contains the highest concentration of GST-GFP for further analysis.

Lab 6: Microplate protein quantification, sample normalization, purification table, and protein crystallization

In Lab 6, students analyze the results from Labs 2–6 to determine the success of their GST-GFP purification. Lysate, affinity, IEX, and flowthrough fraction concentrations are determined in comparison to a BSA standard in a plate reader. Students then normalize the samples in preparation for SDS-PAGE. The data from Labs 2–6 (protein concentrations, volumes, and reaction velocities) are plotted in a purification table, and students calculate specific activity, percent yield of total protein and GST-GFP, and fold purification of GST-GFP. Students analyze their data and determine the success of each step of their protein purification protocol. Students also perform crystallization of lysozyme to demonstrate a technique that can be performed after protein purification.

Lab 7: SDS-PAGE, protein visualization, and western transfer

In Lab 7, students perform SDS-PAGE to analyze their results from Labs 2–6. Students prepare both a resolving and stacking gel, which contain 2,2,2-trichloroethanol (TCE), a chemical that allows for band visualization without the need to fix the gel (as in Coomassie or silver staining). Therefore, only a single gel is required, as students can visualize bands and perform the western transfer using the same gel.

After polymerization, students load their normalized samples, including their original lysate, affinity flowthrough, affinity elution, IEX flowthrough, and IEX elution. They also load a Bio-Rad WesternC marker for visible, chemiluminescent, and fluorescent detection, and GST, GFP, BSA, and lysozyme controls. Gels are run and visualized in a Gel Doc, reviewed with the students, and imaged. Because the samples were normalized, the GST-GFP band (54 kDa) should increase in intensity from the lysate to the affinity and IEX samples, and other nonspecific bands of impurities should be removed as the purification proceeds. GST (26 kDa), GFP (27 kDa), BSA (67 kDa), and lysozyme (14 kDa) should also be visible in the control lanes. Afterwards, gels are transferred onto a

nitrocellulose membrane in preparation for western blotting.

Lab 8: Western blotting

In Lab 8, students perform western blotting on the nitrocellulose membrane to detect GST and GFP. Membranes are blocked and probed with anti-GST and anti-GFP primary antibodies, followed by fluorescently-tagged secondary antibodies. Both proteins are then visualized together with fluorescence. This setup is beneficial as it allows students to see both the GST and GFP channels separately on the same blot, which are then merged, providing valuable information for analysis. Additionally, the fluorescent WesternC marker used in the prior lab is also visible. The GST, GFP, and merged channels are imaged. GST and GFP are visible in their respective positive control lanes (while BSA and lysozyme are negative controls and not visible), and GST-GFP is visible in the lysate, affinity, and IEX lanes. Students analyze their SDS-PAGE and western blot together in a lab report, allowing for a thorough discussion on protein purification, quantification, and analysis. The crystals grown in Lab 6 are also visualized during this lab.

2.3.3 | Section 3: DNA purification, analysis, and recombinant DNA technology

Lab 9: DNA ligation and transformation of E. coli

In Lab 9, students are taught principles on recombinant DNA technology and important vector components such as the antibiotic resistance gene and the multiple cloning site. Students then ligate a GST-GFP DNA fragment into a pGEM-T Easy vector to demonstrate how the gene from the protein section was cloned. This product is transformed into competent *E. coli* cells, which are grown on an agar plate containing ampicillin, IPTG, and X-gal for blue/white screening. Three additional plates act as positive and negative controls for the pGEM-T Easy vector and the competent cells. Blue/white screening results are viewed during the next lab and, if cloning is successful, white colonies will grow on the ligation plate and blue colonies on the vector positive control plate. The competent cells should grow on the control plate with no ampicillin, and the ampicillin plate should have no growth.

Lab 10: DNA purification, DNA quantification, and polymerase chain reaction

In Lab 10, students review the results of the blue/white screening to determine if cloning was successful. Students are provided cell cultures that were grown from blue and white colonies, and perform a miniprep on these cells to purify the pGEM-T Easy plasmid (from the blue cells), and the vector which contains the GST-GFP insert (from the white cells). The purified DNA is then quantified using a Nanodrop or spectrophotometer to obtain the A_{260}, A_{280}, and A_{260}/A_{280} ratio of each sample for concentration and purity calculations. Plasmid-specific primers and a master mix are added to each sample, and PCR is performed. Results are analyzed on an agarose gel in the following lab.

Lab 11: Restriction enzyme digestion, gel electrophoresis, and visualization

In Lab 11, students digest the purified plasmid DNA from the blue and white cell cultures, and analyze the digestion and PCR results on an agarose gel. Students perform a restriction enzyme digestion with EcoRI on the pGEM-T Easy vector (from the blue miniprep) and the vector with the insert (from the white miniprep). Students make an agarose gel and load a DNA marker, their undigested and digested vectors, and their PCR samples. Gels are run and visualized in a Gel Doc, reviewed with the students, and imaged. Both circular and supercoiled DNA are visible in the undigested lanes, and linear DNA in the digested lanes, with bands at 3 kb for the vector, and 1.4 kb for the GST-GFP insert. Additionally, PCR produces a 1.4 kb band from the vector with insert, and a very small fragment from the vector DNA only. Students further analyze the results in their lab report, which allows for a thorough discussion on DNA purification, quantification, and analysis.

2.4 | Laboratory safety

Students are required to wear appropriate clothing and shoes and are provided nitrile gloves, and laboratory coats and glasses when necessary. Protocols contain safety notes in the introduction, and students are informed of proper use of PPE and safety procedures prior to each lab. Safety Data Sheets are utilized to determine safe handling and proper discarding of waste, with oversight from the University's Environmental Health and Safety. The laboratory contains the required signs, has proper sharps and waste disposal containers, and a fume hood. Some reagents, such as Lowry Reagents, Bradford Reagent, CDNB, TEMED, miniprep buffers, and ethidium bromide (ETBR) require special precautions and proper disposal. UV light is used to visualize DNA and proteins, and eyes and skin must be protected from any direct exposure. SDS-PAGE uses acrylamide, and agarose gel electrophoresis uses EtBr, and long pants are required during these labs. The use of laboratory strains of *E. coli*, such as BL21, requires a lab to be designated BSL1 or above.

3 | RESULTS AND ASSESSMENT

3.1 | Student results

In each lab, students record data in their notebook or take pictures of results, which are further analyzed in their lab reports. Sample results are shown in Figures 2–6.

3.2 | Student assessment

Student learning is evaluated through a variety of means, and the laboratory component is worth 25% of the student's course grade (see Table 1).

Before attending the first laboratory, students complete a pre-lab quiz, which reviews the syllabus (see Supplementary Data 3), lab protocols, due dates, academic honesty, and laboratory safety. These topics are further discussed in Lab 1. Attendance is recorded via a sign out sheet at the end of lab. Students take a pre-lab quiz either before lab or in the first 10 min, which is based on prior results and the current lab to help form a connection between the protocols, and to ensure they are prepared for the lab. After lab, students take a post-lab quiz online at home to ensure they can apply the knowledge gained.

Lab progress is further assessed with skill assessments, such as in Lab 1, demonstrating the ability to use a micropipette correctly and filling out a lab report outline. Students also complete a buffer practical which involves calculating amounts of Tris needed, pHing, and making a dilution from a stock solution.

Students analyze their data in a detailed laboratory report, where they demonstrate an understanding and critical analysis of their results. Reports include background information, reagents and equipment, results, and a thorough discussion and analysis of the data, including possible explanations for unexpected results. Reports are graded based on a rubric (see Table 2 for a summary and Supplementary Data 4 for the rubric), and students are provided feedback to improve their critical analysis of their results on subsequent reports. Lab Reports 1, 2, 4/5 (combined), 7/8 (combined), and 11 are full reports, whereas the data from labs 3, 6, 9, and 10 are submitted as results and a short discussion only.

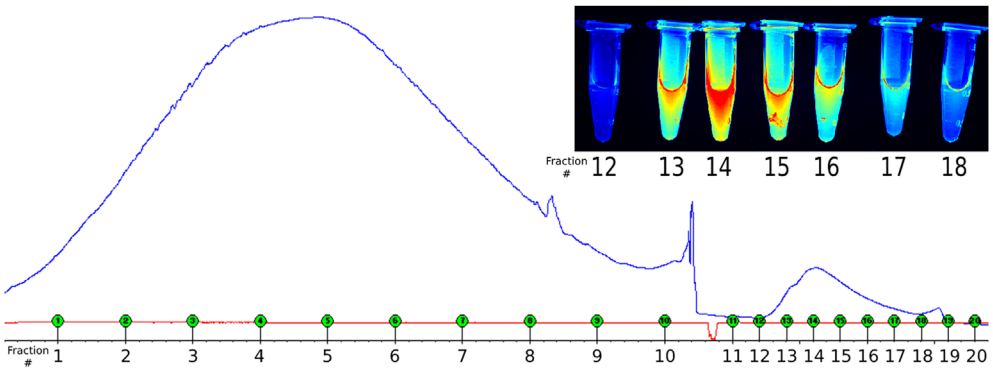

FIGURE 2 Affinity chromatography and fluorescent detection of GFP. In lab 4, fractions 12–18 from the affinity elution (blue line) are placed under UV light to detect GFP fluorescence to locate GST-GFP (inset), which is found at its highest concentration in fraction 14

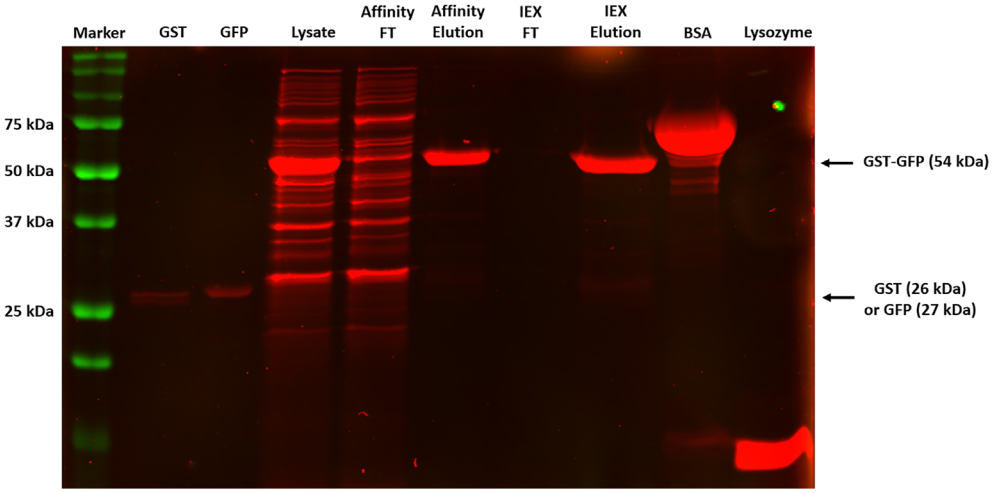

FIGURE 3 SDS-PAGE. Samples from labs 2–6 are run on an acrylamide gel in lab 7. From left to right, lanes contain a marker, the GST and GFP controls, the lysate with expressed GST-GFP (the thick band at 54 kDa), the affinity flowthrough containing bacterial contaminants, affinity elution containing GST-GFP, the IEX flowthrough, the IEX elution containing GST-GFP, and BSA (67 kDa) and lysozyme (14 kDa) controls

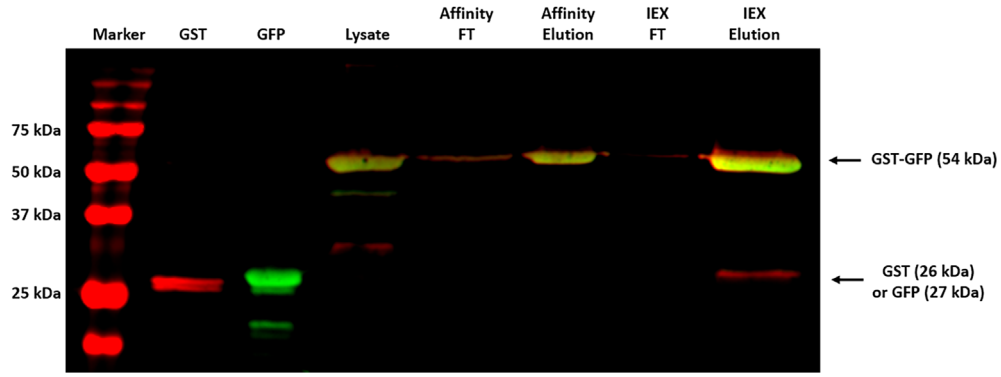

FIGURE 4 Western blotting. The SDS-PAGE gel is analyzed via western blotting in lab 8. Lanes are identical to those in Figure 3. The marker and GST are visible in the red channel, GFP in the green channel, and yellow is visible in the merged image if both GST and GFP are present. The GST-GFP fusion protein is clearly visible in the lysate, affinity elution, and IEX elution lanes as a yellow band

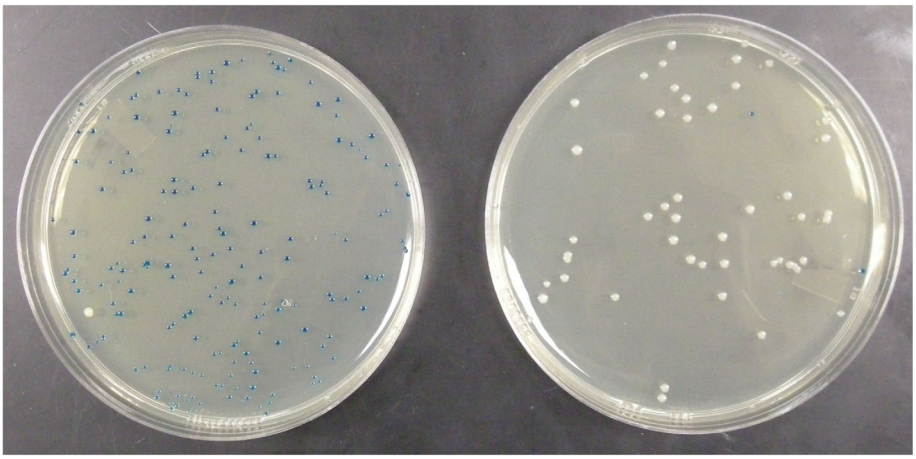

FIGURE 5 Blue/white screening. Ligation and transformation are analyzed via blue/white screening in lab 9. Bacteria transformed with the vector only are blue (left), and bacteria transformed with the vector with the GST-GFP insert are white (right)

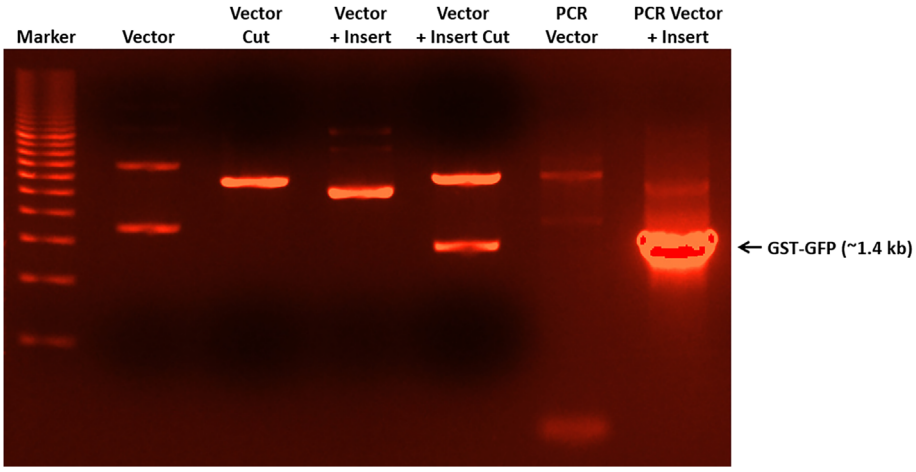

FIGURE 6 Agarose gel electrophoresis. Results from labs 9–10 are analyzed in lab 11 on an agarose gel. From left to right, lanes contain a marker, the vector (3 kb, circular and supercoiled), the digested vector (3 kb linear), the vector with insert (4.4 kb, circular and supercoiled), the digested vector (3 kb) with insert (1.4 kb), PCR of the vector, and PCR of the vector with insert using primers specific to GST-GFP (1.4 kb)

TABLE 1 Point distribution of laboratory course

Item	Number of assignments	Number of points each	% of Total class grade
Pre/Post-Lab Quizzes	22	5	10
Buffer Practical	1	10	
Lab Reports	11	100	15

TABLE 2 Laboratory report rubric summary

Lab report section	% of lab report grade	Requirements
Principle of methods	25	State objective and methods used, describe principles of methods, demonstrate knowledge of the subject
Materials	10	List of reagents (buffers, chemicals) and equipment used in the laboratory
Results	30	Data, tables, pictures, diagrams, and any calculations performed in the laboratory
Discussion	35	State experimental outcomes, critically analyze and explain results, discuss unexpected results

4 | DISCUSSION

The laboratory protocols described herein provide students with hands-on experience with molecular biology research equipment, protocols, and concepts. The sequential nature of the experiments are designed to model a laboratory research project, and students must collect data and critically analyze their results. Protocols have been optimized to provide students with a well-organized laboratory experience that allows for unique results between groups.

Approximately 800 students a year complete the course, in sections of ~25 students, and they work in pairs. Student feedback on the course as a whole (including the lecture section) has been very positive, with student perception of instruction typically above the departmental average regardless of the faculty assigned to the course. Self-reported student comments often commend the laboratory design, organization, critical thinking, and hands-on approach, and rank it highly among labs taken in college. Students have stated that this class brought all of their knowledge from other classes together, and helped to clarify important concepts via its hands-on approach.

This laboratory course has been an important component of our Biomedical Sciences curriculum, and the lab has been continuously improved and optimized over 20 years to provide students with a unique, structured experience. The provided faculty preparation information, student protocols, course syllabus, and rubrics (Supplementary Data 1–4) can be easily adapted to other institutions that teach molecular biology laboratory courses in their degree programs. Additionally, the course can serve as the foundation for an undergraduate research experience, as with our PILOT teaching/research course. We believe this laboratory is readily transferable to other institutions and would be a positive addition to many molecular biology or biochemistry curricula.

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CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest.

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