

**Research-Based Laboratory Promotes Student-Learning Skills and Enhances
Undergraduate Research Experience**

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Abstract

Research experience, at the undergraduate level, is important in understanding the scientific process. However, employment and adaptation of research-based laboratory has been challenging to apply in a large class setting as well as being effective in properly exposing students to core concepts in genetics covered during the lecture. This paper describes how analysis of the *ctrA* gene, which encodes a master cell-cycle transcriptional regulator for bacterial cell-cycle regulation, was adapted and incorporated into the laboratory component of an undergraduate Introductory Genetics course. A total of 159 students participated in this project over a three-semester period, and was carried out with approval from the Institutional Review Board. The test-instrument consisted of collaborative and critical thinking questions dealing with core principles of genetics. Pre-test and post-test scores were compared to assess student-learning and overall effectiveness of the proposed curriculum in light of course objectives. The results validate that the research based laboratory approaches not only broadens student-learning skills their understanding of core concepts in genetics, but also provides the necessary technical skills to carry out independent research projects.

Keywords: Student Learning, Genetics, Undergraduate Research Experience, *ctrA*, Independent Learning

1.0 Introduction

A typical university undergraduate laboratory, across the biology curriculum, consists of students carrying out pre-designed “cook-book” type laboratory exercises. Although the design of this type of lab is simpler and teacher-friendly, student-learning outcomes were found to be minimal through these lab exercises. In addition, due to the repetitive nature of these lab courses, students are often tempted to plagiarize answers from the work of the previous semesters’ students.

As stated by the National Science Foundation, the goal of a college scientific education is to prepare students with core concepts and critical thinking skills that help them in specialized scientific learning and in real-world problems (Brewer and Smith, 2011). Along this line, a major challenge in student learning is how to transform the wealth of information into “critical knowledge” that offers students core concepts and competencies to solve scientific problems. Several different teaching approaches have been employed in the laboratory and lecture to enhance both student learning and their research experiences. Previous studies on the impact of active learning on large biology classes found that it actually increased student learning (Bonwell and Eison, 1991; Ebert-May, Brewer, and Allred, 1997; Udovic, Morris, Dickman, Postlethwait and Wetherwax, 2002).

Biological concepts, without being validated by the student, will remain factual knowledge for them. As such, to promote understanding of these concepts, we have designed a research-based lab approach underlining aspects of the scientific method, whereby laboratory modules were presented in such a way as to guide students into reading research papers, formulating tentative hypotheses, designing and performing experiments, and analyzing results.

This particular lab was designed for an Introductory Genetics course in which students were provided with lab manuals on standard microbiological and molecular experimental methods; however, students were encouraged to use new methods. Students were provided with a list of peer-reviewed scientific articles on a specific topic and required to formulate hypotheses. Following formulation of these hypotheses, students discussed the methods by which they would differentiate the predictions from an alternative hypothesis. However, they were open to conduct independent-research employing alternative, yet feasible, methods that suited their experimental design. This design was intended to provide students with a more realistic experience of scientific methodology. At the conclusion of the lab, students, as a group, were required to collaborate and prepare a cumulative lab report that synthesized the results of all experiments in a “journal article” format.

It is our hypothesis that this proposed laboratory design, that engages students in the research process, will result in a better understanding of the course concepts. The following two hypotheses were tested in this study:

- This research-based laboratory approach enhances student learning.
- Students exposed to the necessary skill-sets and the scientific methodology, through this lab experience, are more independent in the laboratory setting.

2.0 Methods

2.1 Student population, faculty, and teaching assistants

A total of one hundred and fifty-nine undergraduate students, consisting primarily of juniors along with some sophomores and seniors, participated in this study which was conducted over three semesters - Fall 2011, Spring 2012, and Fall 2012 - in the Department of Biological Sciences at Sam Houston State University (SHSU). Differences across pre-test and post-test

results over all three semesters did not show statistically significant differences and hence the data from all three semesters was treated as a single group of analysis. Of these students, 52 (33%) were male and 107 (67%) were female; 95 (60%) were Caucasian, 20 (13%) were African American, 23 (14%) were Hispanic, 5 (3%) were Asian and 16 (10%) were of American-Indian,

	Fall 2011	Spring 2012	Fall 2012	Total
Male	19	18	31	68
Female	32	43	16	91
Caucasian	30	37	28	95
American Indian or Alaska Native	3	0	0	3
Asian	2	1	2	5
Black or African American	6	8	6	20
Hispanic	7	8	8	23
International	2	5	1	8
Unknown	1	2	2	5
No PELL Grant Received	30	40	33	103
PELL Grant Received	21	21	14	56

International, Alaskan or unknown descent, as shown by Table 1.

Table 1. Student demographic data for the duration of the study.

The study was carried out upon approval from the Institutional Review Board (IRB) of SHSU. No significant changes were made to the lab module across the semesters except to the teaching assistants who facilitate to run the laboratory. One faculty member taught the lecture portion of the course across all three semesters. A total of five teaching assistants, of mixed ethnicity and gender, instructed the laboratory each semester and a few were rotated out each semester. Teaching assistants were graduate and senior-undergraduate students who previously took the course and had significant experience with the research methodology employed within this laboratory.

2.2 Research-based laboratory design and lab activities

A research-based approach was employed within the laboratory that complemented the concepts discussed during the lecture portion of the course. More specifically, students were taught in a manner similar to the scientific method whereby they used background information to build hypotheses, design experiments, perform experimental protocols, and analyze results to test the proposed hypotheses.

CtrA is a master cell-cycle transcriptional regulator in *Caulobacter crescentus*, and it regulates coordinated DNA replication and segregation of chromosome during cell division (Dieffenbach, Lowe, and Dveksler, 1993). The *ctrA* gene has been identified in other proteobacterial species, however, its function has not yet been determined. As such, students were provided with a list of peer-reviewed scientific articles on the Bacterial cell cycle and the cell-cycle transcriptional regulator (*ctrA*) to utilize as resources to gain sufficient background to identify the scientific problem and formulate two-to-three tentative hypotheses to be tested in this laboratory. Furthermore, they were required to read and review the information prior to the first day of laboratory and write important background information.

On the first day of laboratory, students were randomly assigned into groups of three. Within each of these groups, students were required to actively participate and discuss the reading assignments, a process facilitated by the teaching assistants. The instructor would frequently propose questions to not only invoke critical thinking and collaborative learning skills, but also to aid in the formulation of hypotheses. Collaborative learning skills are those in which the students work in a group towards a common goal; critical thinking skills are those that require the students to analyze, synthesize, and evaluate core concepts (De Hei, 2014). Using a guided question-answer session as a template, students formulated three correlated hypotheses.

The following three hypotheses were formulated by the students:

(1) The *ctrA* gene is present in bacterial species tested, including *Rhodobacter sphaeroides* 2.4.1, *Rhodobacter sphaeroides* 17029, *Rhodopseudomonas palustris* BisB5, and *Escherichia*.

(2) As *ctrA* gene is essential for the cell cycle, sequences of *ctrA* genes isolated from different species are highly conserved.

(3) As *ctrA* gene regulates its own gene expression, *ctrA* genes of different species do share common regulatory sequences in their promoters.

Following formulation of these three hypotheses, students then discussed the methods by which they would differentiate the predictions from an alternative hypothesis. For example, the first hypothesis predicts that the *ctrA* gene is present in all bacterial species. Application of the PCR method would amplify *ctrA* genes from species representative of different groups of bacteria. Consequently, if the student was able to obtain PCR product from all species examined it would support the first hypothesis. Selected methods were subsequently aligned to each hypothesis and predicted results were discussed within each group. These methods included, but were not limited to, primer design, genomic DNA preparations, polymerase chain reaction (PCR), analysis of PCR products through gel-electrophoresis, DNA sequencing and DNA and protein sequence analyses using NCBI associated tools available on publically available websites.

During the experiments, the lab instructor would identify to the student's equipment and purposes of specific steps in the protocol, to enhance their understanding of not only the equipment and reagents used in the experiment but also the scientific process. The students were then required to apply these methods, interpret the results and take the detailed notes in their laboratory notebooks. At the conclusion of the lab, students, as a group, were required to

collaboratively prepare a cumulative lab report that analyzed the results from all of the experiments in “journal article” format. More specifically, within the group, each student was assigned to write specific sections of the lab report. Following completion of each section the students then evaluate each other’s portion and suggested specific comments for revision. Upon successful revision, the students then compiled the sections into a complete manuscript, following the guidelines set forth by the rubric, for submission as the final lab-report. Furthermore, a table presenting an overview of weekly lab activities is provided in Table 2.

Table 2. Weekly activities for the designated laboratory.

Week 1	Introduction of scientific process, discussion on background papers, proposing tentative hypotheses, and primer design
Week 2	Preparation of Genomic DNA and alignment of hypothesis with specific experimental design
Week 3	Performing polymerase chain reaction (PCR) and discussion on hypotheses
Week 4	Purification of PCR products and DNA sequencing
Week 5	DNA sequence analysis: Identification of <i>ctrA</i> gene and performing codon analysis
Week 6	Protein sequence analysis: Amino acid analysis and motif structure analysis
Week 7	Analysis of promoter and termination sequences
Week 8	Draft paper: Peer-review session, providing suggestion on manuscript

2.3 Pre-lab and post-lab assessments

The pre-test and post-test instruments were 50 multiple-choice type questions consisting of 25 collaborative learning (CL), 20 critical thinking (CT), and 5 control questions. Questions of the control category were taken from unrelated topics that were not covered either in the lecture or laboratory of this course. Furthermore, pre- and post- tests were conducted on the first and last days of the laboratory, respectively. The scores were then analyzed for student progress within the two main areas of active learning: collaborative learning and critical thinking. This test was an indicator of the students overall improvement in the lab and understanding of

molecular concepts. In addition, lab reports were evaluated based on a rubric provided to students prior to the manuscript preparation.

An identical test was given for all three semesters. Students who took both pre- and post-tests were confidentially coded for data storage and analysis. The data on student demography, gender, and socio-economic status were provided by the Office of Institutional Effectiveness at Sam Houston State University.

2.4 Statistical analysis

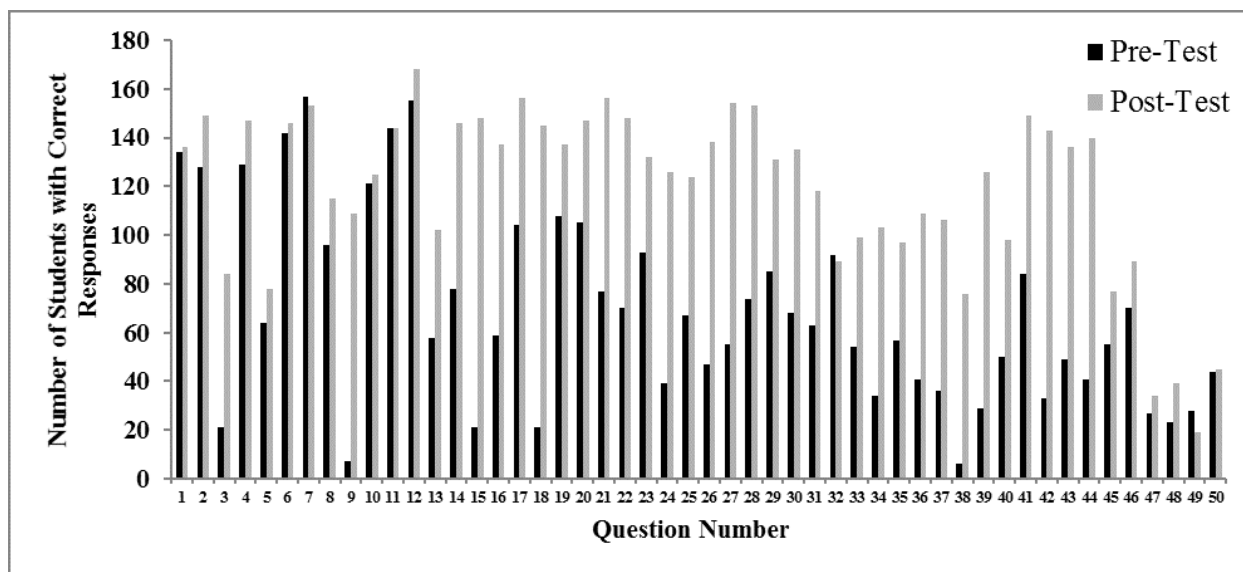
The quantitative data includes the number of correct and incorrect responses for all fifty questions. Paired t-tests were performed to measure the students' collaborative learning and critical thinking skills and their knowledge of the conceptual contents. Chi-square (χ^2) analysis was employed to test the number of undergraduate students enrolled in independent research projects with a null hypothesis assuming that there was no significant increase in enrollment from 2009-2010 to 2011-2012.

3.0 Results and Discussion

3.1 Research-based laboratory promotes active learning

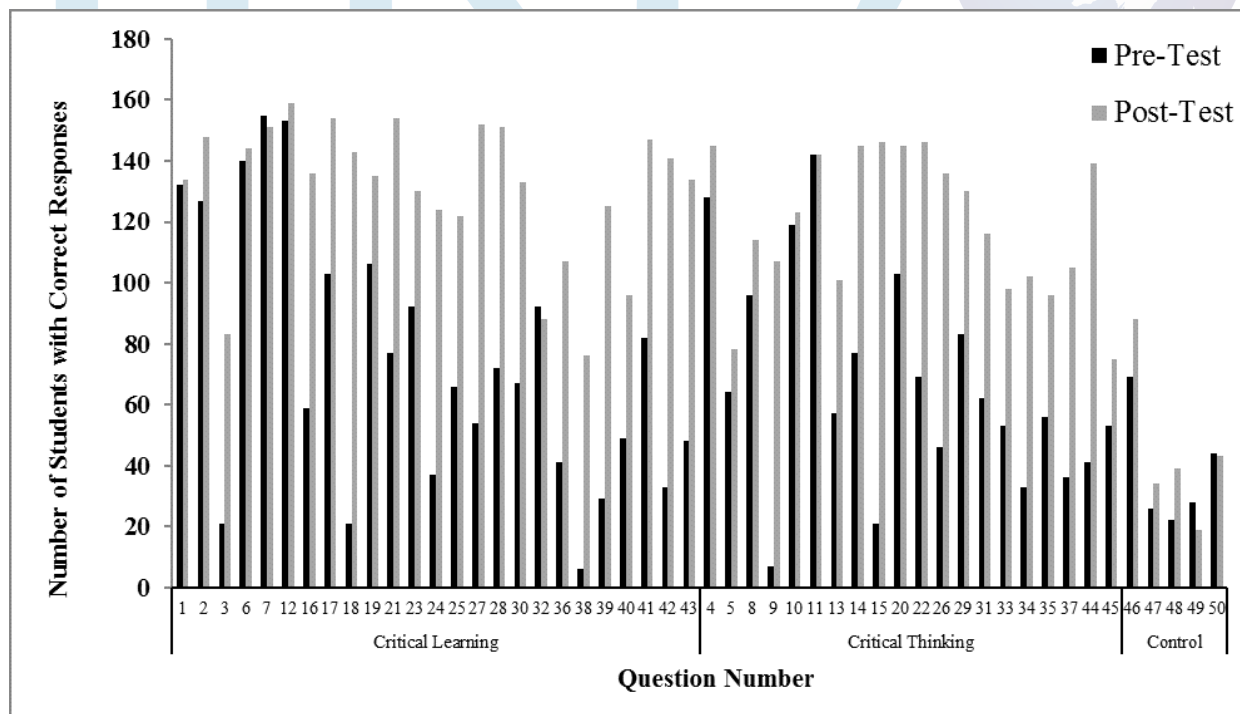
Since data for each individual semester had similar trends, analyses were performed and reported as one integrated study. The number of correct responses to each question in the post-test was significantly greater when compared to the pre-test as shown by Figure 1.

Figure 1. Number of correct student responses per question.



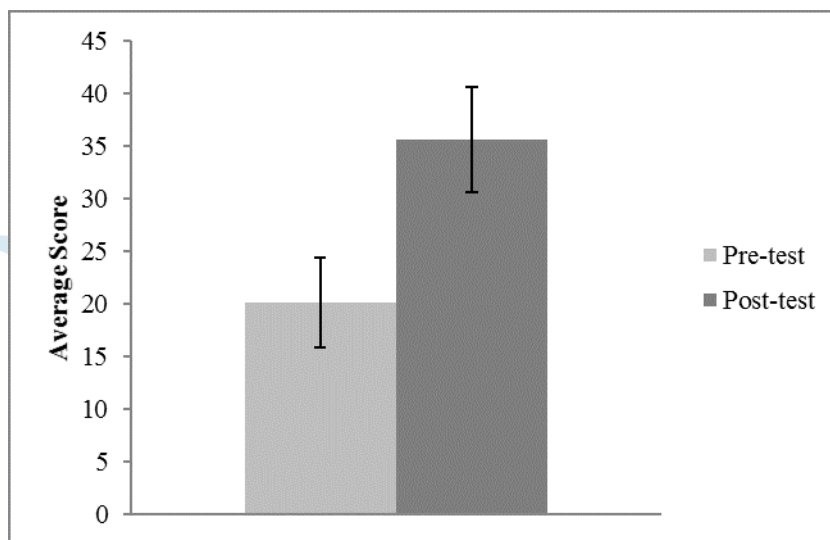
The questions were analyzed based on the types of questions, as shown in Figure 2.

Figure 2. Number of correct student responses per question separated by type of question.

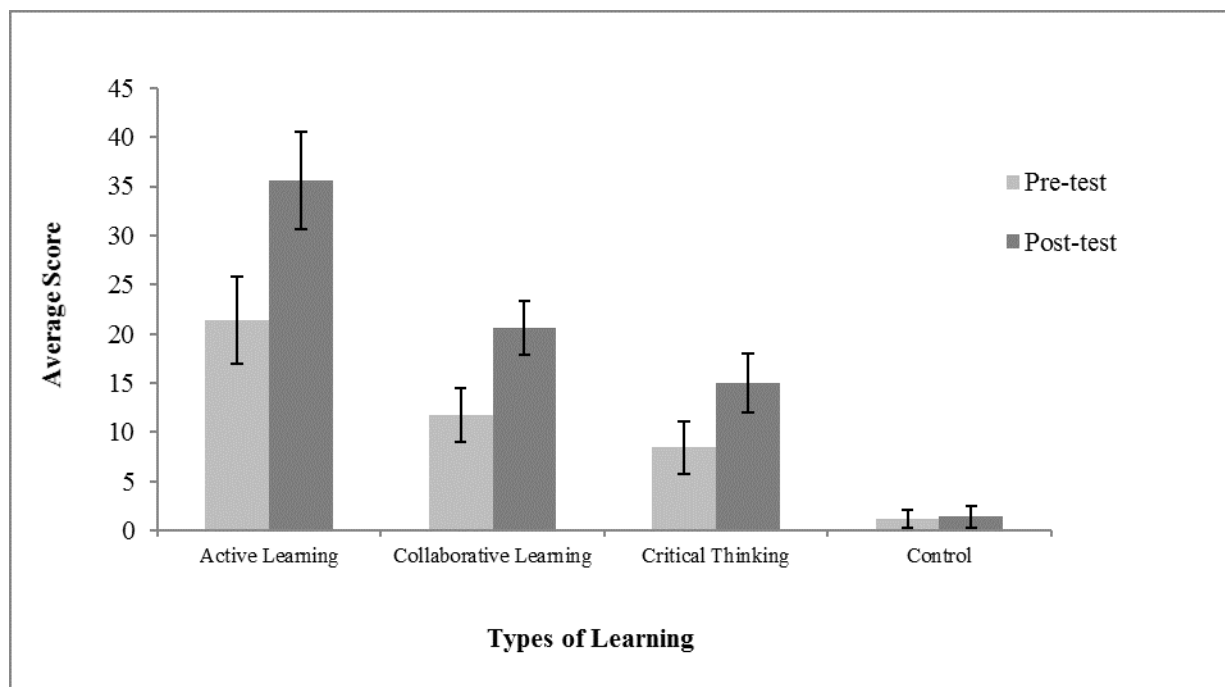


Nearly all questions were answered correctly by more students in post-test than in pre-test, with the exception of question numbers 7 and 32, for which the numbers of correct responses in pre-test were slightly greater. Both of these questions were collaborative-type learning questions, and may not have been fully discussed during the collaborative, group activity.

Figure 3. Average pre-test and post-test scores.



The average test score, as shown by Figure 3, significantly increased from 26.31 ± 4.27 (pre-test) to 41.83 ± 3.72 (post-test). Improvements not specifically outlined by the study but noticed by the instructor and the teaching assistants were that the students were much more confident in their knowledge when the lab was based on scientific method as compared to the traditional approach employed during previous semesters. When test scores were analyzed by question-type (collaborative, critical thinking, and control), students showed significant improvement in both collaborative and critical thinking but not in the control, as shown in Figure 4.

Figure 4. Average pre-test and post-test scores for different types of learning.

The average pre-test scores, post-test scores and p-values are provided in Table 3.

Table 3. Average pre-test and post-test scores for different types of learning.

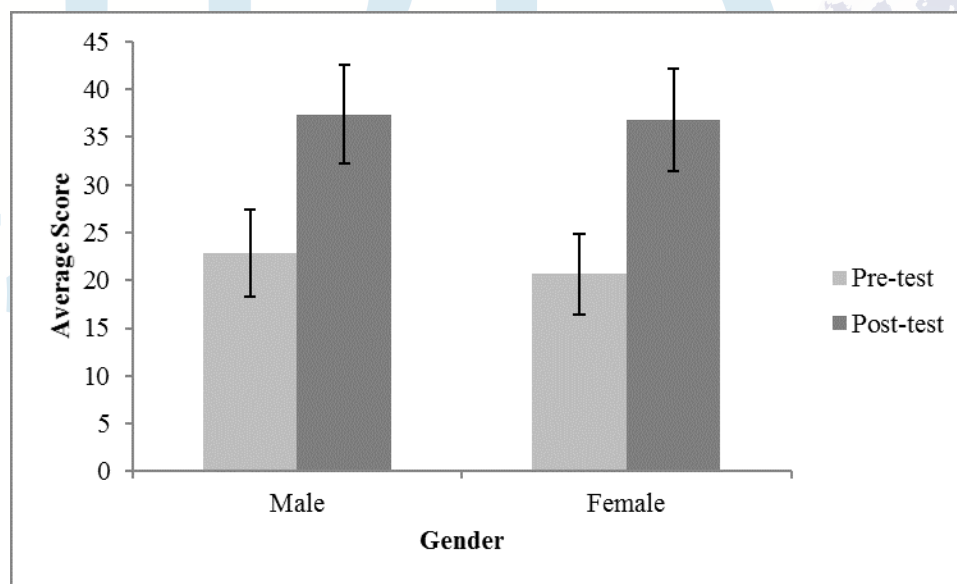
	Pre-Test	Post-Test	P-value
Active Learning	20.107±4.241	35.616±4.947	2.878E-65
Collaborative Learning	11.710 ±2.738	20.591±2.693	1.407E-66
Critical Thinking	8.465±2.678	15.025±2.987	2.795E-48
Negative Control Group	1.188±0.942	1.402±1.097	1.041E-2
Total	26.31±4.27	41.83±3.72	

Scores for collaborative and critical learning questions increased from 11.71 ± 2.74 to 20.59 ± 2.69 and from 8.47 ± 2.68 to 15.03 ± 2.99 , respectively, demonstrating that student learning significantly increased. There were five questions in the control group that were not discussed in the laboratory, constituting an internal negative control group. The number of correct responses to the questions of the control group slightly increased from 1.18 ± 0.94 to 1.40 ± 1.10 , (p value = 0.01), which indicates a significant increase under conventional criteria, but the strengths of

the p-values for both collaborative and critical thinking were several orders of magnitude lower than the levels of p-values observed for the control group pre- and post-test comparison. Taking into account that there were only a limited number of five questions within the control group, it can be safely disregarded, and has no impact on the results of a research-based approach found within this study. However, it is also quite possible that these questions may have been discussed in other related biology courses and that the students had prior knowledge about these questions.

3.2 No significant difference in learning outcomes between genders, socio-economic groups and ethnicities

Figure 5. Average pre-test and post-test scores for male and females.



Students were categorized by gender, ethnicity and socio-economic status as shown in Table 1.

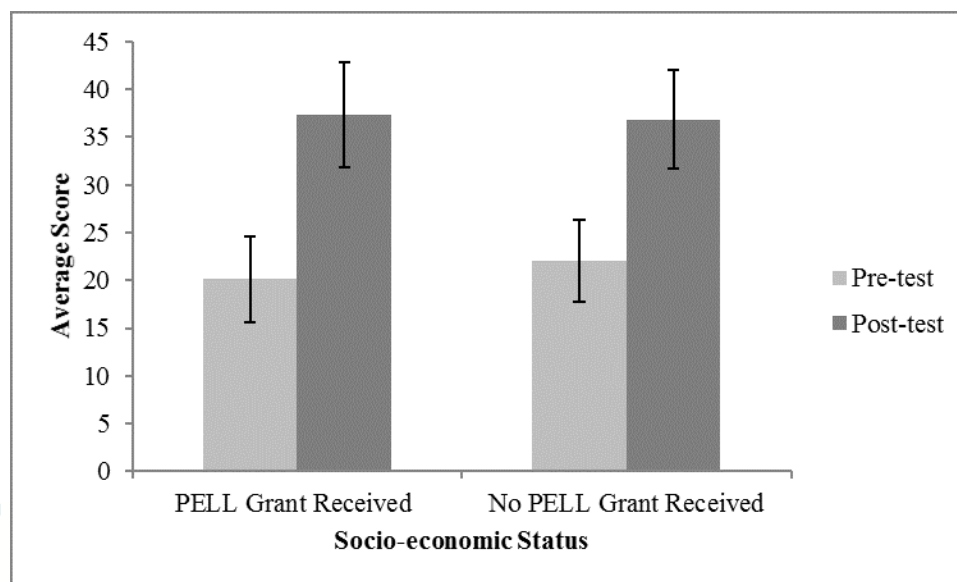
The average pre-test scores, post-test scores, and their corresponding p-values were calculated independently for each category as shown in Table 4.

Table 4. Average pre-test and post-test scores for different demographics.

	Pre-Test	Post-Test	(P-value)
Male	22.846 ±4.552	37.403 ±5.149	3.978E-42
Female	20.644 ±4.245	36.813 ±5.355	2.405E-54
Caucasian	22.231±3.969	37.736 ±4.957	4.395E-52
American Indian or Alaska Native	17.666 ±4.041	44.000 ±2.645	3.555E-57
Asian	20.600 ±5.683	38.800 ±5.890	1.978E-32
Black or African American	19.000 ±4.790	34.350 ±5.724	2.377E-44
Hispanic	20.869 ±4.693	35.608 ±5.441	3.928E-42
International	19.875 ±5.718	36.250 ±3.882	4.111E-54
Unknown	22.000 ±4.795	35.400 ±6.024	2.231E-31
No PELL Grant Received	22.038 ±4.295	36.825 ±5.185	2.117E-42
PELL Grant Received	20.125 ±4.516	37.339 ±5.481	1.401E-46

Males had average pre-test and post-test scores of 22.85 ± 4.55 and 37.40 ± 5.15 , respectively. Similarly, females had average pre-test and post-test scores of 20.64 ± 4.25 and 36.81 ± 5.36 , respectively. An unpaired t-test was performed between post-test scores of males and females to ascertain whether any significant difference in performance existed by gender. The comparison of students' post-test scores between males and females were found not to be significantly different ($p = 0.5098$).

Figure 6. Average pre-test and post-test scores per socio-economic status.



Socio-economic status was defined by whether a PELL grant was received by the student or not. For those who received a PELL grant the average pre- and post- test scores were 20.12 ± 4.52 and 37.34 ± 5.48 , respectively, while for those who did not receive PELL grant, pre- and post-test scores were 22.04 ± 4.30 and 36.83 ± 5.19 , respectively as seen in Figure 6. To ascertain whether there was a significant difference in performance by socio-economic status, an unpaired t-test was performed between the post-test scores received by students from those two categories. Results indicated no significant difference ($p = 0.559$) between the pre and post-test scores of these two economic groups.

Comparisons of average pre- and post-test scores of all categories of ethnicity, as shown in Table 4, demonstrated significant increase in their post-test scores. Furthermore, no significant differences in student performance between ethnicities were noted upon analysis. The ethnicity was analyzed as Caucasian and non-Caucasian to accommodate the difference in the sample size

of each ethnic group. -For continued research, it was interesting to note that the class sizes were predominantly Caucasian (60%) and hence an interesting point of continuance for this study would be to see whether greater number of minority students alters the trends found within the current study.

4.0 Conclusion

Overall, comparison of pre- and post- test scores show that student understanding of Introductory Genetics improved significantly by the application of the research-based approach, designed in this study. This was further supported when the test questions were divided into their respective types of learning - collaborative learning and critical thinking - whereby students had statistically significant increases in both types of active learning skills. These results agree with previous studies that show the effectiveness of active learning in improving student learning skills (Bonwell et al. 1991; Ebert-May et al., 1997; Udovic et al., 2002). This approach also increased the number of students enrolled in independent research studies over the course of time that this study was employed. The authors note that this laboratory experience is not the only possible causation of the increase in the enrollment within independent research, but feel as though the experience lead to an increase in the student's capabilities. The design of the laboratory sessions allows for independent thinking and problem solving through many activities. Students are expected to carry out experiments independently within the laboratory, including the troubleshooting of the primer design, gel electrophoresis techniques, and PCR protocols. Students are also expected to draft a manuscript of the results of the laboratory experiments performed. These types of activities strengthen the independent learning that students require when moving into independent research studies.

All aspects of this study exemplify that a research-based laboratory both promotes student learning and enhances undergraduate research experience, and thus can be successfully applied not only to genetics laboratory but also to a number of other biology laboratory courses.

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