

Method to Increase Undergraduate Laboratory Student Confidence in Performing Independent Research[†]

Colton E. Kempton, K. Scott Weber, and Steven M. Johnson*

Department of Microbiology and Molecular Biology, Brigham Young University, Provo, UT 84602

INTRODUCTION

Cookbook-type protocols, common to high school and undergraduate-level science classes, are a less effective means of instruction than inquiry-based labs as they allow students to be passive and typically do not require critical thinking (1). They do not accurately reflect the investigative nature of science, where there is no accompanying fill-in-the-blank, universal protocol that is used to discover new information (2, 3). To improve the quality of science education, there has been a push to replace these cookbook-style protocols with more open-ended investigative or inquiry-type instruction that is student centered (4–7). Our research has demonstrated increased engagement when students use, or anticipate using, data from their own genome (8). Inquiry-based learning activities model the scientific process much better than cookbook labs and lead to increased understanding of the scientific process (9, 10).

To promote independent learning, we designed our undergraduate course to apply the scaffolding instructional methodology (11) to wean students from cookbook laboratory procedures by sequentially introducing protocols with decreasing amounts of written instructor guidance. Scaffolding originated as adults helped children develop higher psychological functioning and ability to express themselves through guided interactions (12, 13), ultimately enabling children to do things independently that normally require adult guidance and assistance (13). We have applied this method to our undergraduate Advanced Molecular Biology Laboratory at Brigham Young University (Appendix I, Methods) with the goal of teaching the students to find and use protocols and develop scientific independence. This method enables student transition from instructor dependence to scientific independence.

General application of this method involves students performing a series of planned experiments while sequentially

providing them with 1) protocols with step-by-step instructions typed out by the professor, 2) instruction with manufacturers' protocols augmented with additional explanations inserted by the professor, 3) unaugmented manufacturers' protocols, 4) protocols received from scientists, 5) a primary literature protocol, and finally, 6) protocols found by the students themselves (Fig. 1). We applied this method to our Advanced Molecular Biology Laboratory course. Results from our student survey demonstrated significant increases in student confidence to use and adapt new protocols to carry out experiments. Students also showed greatly increased confidence in their ability to troubleshoot and to carry out independent research experiments.

PROCEDURE

Simple, professor-provided protocols

We start with simple, professor-written protocols (Fig. 1). These instructions include detailed steps to accomplish the experiments adapted from kit instructions and simplified for ease of use.

We applied this principle with our DNA fingerprinting module (Fig. 2): students isolate genomic DNA (14), perform PCR, do PCR DNA cleanup and restriction enzyme digests (15), and analyze DNA on gels. Each of the protocols is step-by-step instructions typed out by the professor.

Manufacturers' protocols with added instructions

In the second phase we use protocols/instructions that come with kits, supplemented with additional instructions by the professor (Fig. 1).

Our Site-Directed Mutagenesis module (Fig. 2) applies this principle. Students isolate plasmid DNA, perform site-directed mutagenesis, bacterial transformations, colony selection, and colony PCR, and sequence PCR products. We use supplemented protocols from the QIAprep Miniprep (16), PHUSION Site-Directed Mutagenesis (17), and the ZERO BLUNT TOPO PCR Cloning Kits (18). Students apply first module protocols as they perform restriction digests, gel electrophoresis, and colony PCR in preparation for sequencing to confirm the success of their mutagenesis.

*Corresponding author. Mailing address: Brigham Young University, 4007 LSB, Provo, UT 84602. Phone: 801-422-9170. Fax: 801-422-0004. E-mail: stevenj@byu.edu.

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Manufacturers' protocols

In the third phase we provide students with only the protocols/instructions that come from the kit (Fig. 1). We use three protocols (short, long, and average-length) students might actually experience in the real world. Students must glean what is necessary from the protocol to be able to do the experiment.

Our Northern Blotting module applies this principle (Fig. 2). Students isolate RNA using TRIZOL Reagent (19), with a two-page protocol outlining multiple procedures. This is followed by northern blotting using a detailed 42-page NORTHERNMAX-GLY kit and protocol (20). Students look through the protocol and decide which steps to include for

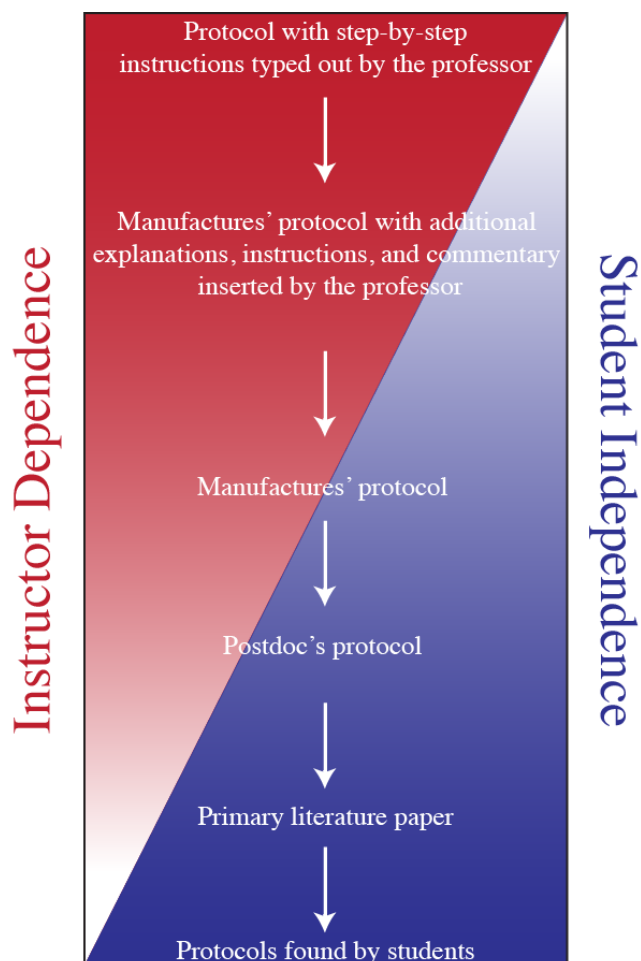


FIGURE 1. Overview of the application of the weaning philosophy and approach. The weaning approach is applied to any laboratory class by initially providing students with protocols that are highly modified by the professor (1st and 2nd), followed by protocols with decreasing amounts of professor modifications and protocols with no professor modifications (3rd), and finally resources from which the students must extrapolate protocols (4th and 5th). Ultimately, students are not provided with protocols, but instead find protocols on their own (last). The red color on the left that decreases from top to bottom represents the amount of student dependence on the written instructions from the professor, and the blue color on the right that increases from top to bottom represents the amount of student independence at each stage of the weaning.

their application. Finally, we use the Chemiluminescent Nucleic Acid Detection Module and protocol, a straightforward kit and instructions, to visualize the probe on their blots.

Real-life protocols

The final phase in our methodology toward independence is to use protocols the students might receive from other researchers when trying to reproduce published techniques. Students receive a protocol sent from a postdoctoral fellow and a primary-literature paper from which they need to reproduce an experiment. Students follow the postdoc protocol and read and understand the primary literature paper to glean what they need to replicate the experiments contained therein. These are the types of protocols they might encounter in a research career. Using and applying them in a carefully controlled laboratory experience prepares them for independent research.

We applied this principle with our Electroporation Mobility Shift Assay (EMSA) module. The instructional resources for these experiments are a primary research paper (21) and a protocol from a postdoc (G. Ramaswamy, 2003. Gopi's nuclear extract protocol, personally communicated protocol to isolate nuclear proteins from *C. elegans*). Students determine how to perform the EMSA in the paper from the materials and methods section and additional outside resources online. The terse protocol provided by the postdoc has each step for nuclear protein isolation, but no logistical commentary. The chemiluminescence kit and protocol used in the third module is again used here to reinforce the skills they previously acquired.

Independent application

Having experienced a range of instructional materials and performed several molecular techniques, students are asked to directly apply what they have learned throughout the semester. The culminating event is when students choose, design, and perform independent projects for the last four weeks of the semester. Students independently come up with their own scientific questions, plan the procedures, find the necessary protocols, and perform the experiments. Instructors only approve their projects and provide the necessary reagents.

The pinnacle event is the last day of class when students present their independent projects, complete with background, hypothesis, experimental procedures, data, results, and conclusions to the entire class. With the final independent project, the students have moved from preplanned, instructor-dependent, results-controlled experiments to independently conceived, designed, and executed projects that succeed or fail based on the student.

CONCLUSION

Here we present the application of a scaffolding pedagogical method to transform undergraduate laboratory students into independent researchers. We surveyed student

Module	Experiment	Resource
DNA Fingerprinting	DNA isolation PCR PCR DNA clean-up Agarose gel electrophoresis Restriction enzyme digestion	Protocol and step-by-step instructions typed out by the professor
Site-directed Mutagenesis	Plasmid isolation Restriction digest and electrophoresis Site-directed mutagenesis Bacterial transformation Colony selection and colony PCR DNA sequencing submission & analysis	Manufactures' protocol with additional explanations, instructions and commentary inserted by the professor
Northern Blotting	RNA isolation Northern blotting Chemiluminescent detection	Manufactures' protocol
EMSA	Nuclear protein isolation Electrophoretic mobility shift assay Chemiluminescent detection	Postdoc's protocol Primary literature paper
Independent Projects	Variable	Protocols found by students

FIGURE 2. Specific application of the weaning philosophy and approach. The name of each specific module (left) is listed with its accompanying experiments (middle) and the type of resources that are provided for those experiments (right). The color and intensity of the background fields of the modules represent the amount of professor dependence (red) or student independence (blue) in each module (see Fig. 1). Experiments listed in green print are procedures that the students have learned in previous modules. The other color print (black or white) differs only for ease of reading.

attitudes about their abilities to perform independent research. Student abilities to independently plan and execute appropriate experiments increased, as did their confidence to do independent research (Appendix 2, Measuring Learning). This methodology is likely applicable to any lab course in life sciences striving to develop independent undergraduate researchers. Consistent results between three sections taught by three different professors suggest that this method is not instructor specific, but generally applicable.

SUPPLEMENTAL MATERIALS

- Appendix 1: Methods (course sections, survey instrument, data analysis, advanced molecular biology laboratory protocols, laboratory safety procedures)
- Appendix 2: Measuring learning and supplemental figures (S1, S2)

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REFERENCES

1. Modell HI, Michael JA. 1993. Promoting active learning in the life-science classroom—defining the issues. *Ann New York Acad Sci* 701:1–7.
2. Cox DD, Davis LV. 1972. The context of biological education: the case for change. Commission on Undergraduate Education in Biological Sciences, Washington, DC.
3. McComas WF. 1998. The nature of science in science education: rationales and strategies. Kluwer Academic Publishers, Dordrecht, Netherlands.
4. Boyer Commission on Educating Undergraduates in the Research University. 1998. Reinventing undergraduate education—a blueprint for America's research universities. State University of New York at Stony Brook for the Carnegie Foundation for the Advancement of Teaching, Stony Brook, NY.
5. National Research Council (USA). Committee on Undergraduate Biology Education to Prepare Research Scientists for the 21st Century. 2003. Bio 2010: transforming undergraduate education for future research biologists. The National Academies Press, Washington, DC.
6. National Research Council (USA). Committee on Development of an Addendum to the National Science Education Standards on Scientific Inquiry. 2000. Inquiry and the National science education standards: a guide for teaching and learning. The National Academy Press, Washington, DC.
7. Woodin T, Carter VC, Fletcher L. 2010. Vision and Change in Biology Undergraduate Education, A Call for Action—Initial Responses. *CBE Life Sci Educ* 9:71–73.
8. Weber KS, Jensen JL, Johnson SM. 2015. Anticipation of personal genomics data enhances interest and learning environment in genomics and molecular biology undergraduate courses. *PLoS One* 10:e0133486.
9. DiPasquale DM, Mason CL, Kolkhorst FW. 2003. Exercise in inquiry. *J Coll Sci Teach* 32:388–393.
10. Handelsman J, Ebert-May D, Beichner R, Bruns P, Chang A, DeHaan R, Gentile J, Lauffer S, Stewart J, Tilghman SM, Wood WB. 2004. Education. Scientific teaching. *Science* 304:521–522.

11. Beed PL, Hawkins EM, Roller CM. 1991. Moving learners toward independence—the power of scaffolded instruction. *Read Teach* 44:648–655.
12. Bruner JS. 1975. The ontogenesis of speech acts. *J Child Lang* 2:1–19.
13. Vygotsky LS. 1978. *Mind in society: the development of higher psychological processes*. Harvard University Press, Cambridge, MA.
14. Invitrogen. 2007. PureLink™ Genomic DNA Kits, for purification of genomic DNA, User Manual, Version A. Life Technologies Corporation, Carlsbad, CA.
15. Invitrogen. 2010. PureLink® quick gel extraction and PCR purification combo kit, for purification of DNA fragments from agarose gels and rapid, efficient purification of PCR products, user manual, MAN0001636. Life Technologies Corporation, Carlsbad, CA.
16. Qiagen. 2002. QIAprep® Miniprep handbook, second ed. Qiagen, Hilden, Germany.
17. Finnzymes. 2008. Phusion™ site-directed mutagenesis kit, instructions, Version 1.1. New England Biolabs, Inc., Ipswich, MA.
18. Invitrogen. 1999. Zero Blunt® TOPO® PCR cloning kit, five-minute cloning of blunt-end PCR products, user manual, version N. Invitrogen Corporation, Carlsbad, CA.
19. Ambion. 2007. TRIzol® reagent, instructions. Life Technologies Corporation, Carlsbad, CA.
20. Ambion. 2008. Instruction manual, NorthernMax®-Gly, Glyoxal-based system for northern blots, P/N 1946M Revision B. Applied Biosystems, Carlsbad, CA.
21. Johnson SM, Lin SY, Slack FJ. 2003. The time of appearance of the *C. elegans* let-7 microRNA is transcriptionally controlled utilizing a temporal regulatory element in its promoter. *Dev Biol* 259:364–379.