A Multi-Week Inquiry for an Undergraduate Introductory Biology Laboratory

Investigating Correlations Between Environmental Variables and Leaf Stomata Density

Bruce W. Grant and Itzick Vatnick

Numerous authors have noted that open-ended inquiries give students a sense of ownership of their learning and that this contributes to their development as life-long learners. We wish to add that teaching using these types of activities promotes the same goal in teachers as well.

In the last decade, undergraduate science education has undergone a quiet revolution emphasizing opportunities for student-directed original inquiry *as* the curriculum.

Opportunities to conduct open-ended investigations in the laboratory portion of courses are important because they teach students how science is actually done and thereby they learn by doing it (Woodhull-McNeal 1989; Goodwin et al. 1991; Heady 1993; Ortez 1994; Sundberg and Moncada 1994).

However, several authors have commented that biology departments seem to have lagged behind in these visionary reform efforts of course curricula for a variety of reasons (Holt et al. 1969; Carter et al. 1990; Sundberg and Armstrong 1993), and our experience in teaching introductory labs leads us to concur.

Our explanation is that many of our freshmen students lack sufficient confidence, organizational skills, and content background for open-ended student-directed investigations *as freshmen*, and we lack the resources to supervise large numbers of independently active learners that populate our introductory courses. Our solution is to begin with an intermediate step that retains at least some of the ownership and empowerment components of open-ended inquiries as well as the efficiencies of scale of the "demonstra-

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Table 1: Inquiry Framework. Summary of the relations among various modes of inquiry for introductory laboratory courses in biology. Columns refer to various components of the inquiry, and the cells indicate who "owns" each component, i.e., was the component provided by the instructor or up to the student to generate? (modified from Sundberg and Moncada 1994; Ohlhorst 1995; D'Avanzo 1996)

inquiry mode	research question	study system	data collection/ statistical analysis	data interpretation	results and conclusions
open-ended inquiry	student	student	student	student	student
bounded inquiry	student/ given	student	student/given	student	student
guided inquiry	given	given	given	student	student
closed-ended demonstration	given	given	given	given	given

tion lab" format. We refer to this intermediate as a "bounded inquiry."

In bounded inquiries, students' research questions and study subjects are instructor constrained but hypotheses and study species are not a priori specified. In addition, sample acquisition, data collection, statistical analyses, and other technical details are prescribed, however, neither the students nor the instructor know what the outcome of the study will be. Finally, a critical component of this activity is that at the end, students present their research findings to their peers during an in-class research symposium using their own authentic voices. A summary table showing the relations among the various inquiry modes appears in Table 1.

We have developed a bounded inquiry for the second semester laboratory of our two-semester first-year biology course. This inquiry involves the study of the ecophysiology of terrestrial plants and is titled "Environmental Correlates with Leaf Stomata Density." During the inquiry, students investigate correlations between various environmental variables (light, temperature, carbon dioxide, and so forth) and leaf stomata density. Below, we describe the salient features of the bounded inquiry our students conducted, and we relate some of the student results from and attitudes toward this inquiry over the past three years.

Synopsis of Lab Activity

Lab Period #1:

(a) We perform an introductory activity during which instructors use a combination of lecture and group questioning to teach the basic ecophysiology of leaf stomata including stomata structure, function and role in the regulation of leaf temperature, gas exchange, and photosynthesis in vascular plants (our lab handout is available on a course-specific web page linked to the Widener University biology department web site: http://www.science.widener.edu/~grant/esa/exp2/bgiv_1.html).

(b) Students work in pairs to envision a specific environmental difference that might affect stomata density, and formulate predictions about which way they would expect stomata density to vary. At this stage students are asked to generate a graph that depicts what their data would look like if their predictions were (or were not) true. The environmental difference of interest to them must be available on campus, and before they head out they must envision exactly where to go to obtain leaves for their study. All of the above must be discussed with and approved by the instructor PRIOR to collecting data.

(c) Students bring their leaf samples back to lab and estimate their stomata densities using a clever technique of making stomata impressions using commonly available nail polish and clear tape as is described in **Appendix 1** (Neill et al. 1990; Brewer 1992; see http://www.zoo.toronto.edu/zooweb/able/volumes/vol-13/3-brewer/3-brewer.htm). Stomata impressions are easily visible under a light microscope at 400x (see **Figure 1**).

Lab Period #2:

(a) Students finish counting their stomata slides, if they have not yet done so.

(b) Instructors use a combination of lecture and group questioning to teach basics of statistical analysis that students will need to analyze their data (including averages, standard deviations, statistical terminology [e.g., significance level, P value, degrees of freedom], and the use of a t-test).

(c) Instructors demonstrate the use of a simple spreadsheet and statistics package (Microsoft's EXCEL) and help students to input their data, calculate averages and standard deviations, perform a t-test using these data, and generate a clear graphic summarizing their results.

In actuality, only half of lab period #2 is devoted to the activities listed above. In the first half of lab, students perform an unrelated and more traditionally formatted lab on fruit and flower diversity. Between lab periods 2 and 3, students generate coauthored oral and written reports according to specific guidelines (included with the lab handout mentioned above).

Lab Period #3:

Each pair of students presents their data and conclusions in class in a 10-minute report to their peers during the "Stomata Results Symposium."

SUMMARY OF RESULTS

During the spring semesters of 1995, 1996, and 1997 we supervised over 75 projects, all of which used green plant material available in mid-February on Widener's campus (this happened despite the fact that Widener is located in an urban landscape and in 1996 a foot of snow was on the ground).

The majority of the projects were studies of the effects of sunny versus shady environments on stomata density. About half of these compared different individuals of the same species living in different places on Widener's campus, and the other half examined leaves in the sun versus shade on the same individual plant (e.g., within a dense shrub or tree). The remainder of the projects examined other effects such as proximity to sources of carbon dioxide (the campus is adjacent to interstate 95) and the effects on grass of trampling by pedestrians.

The vast majority of the results were consistent with the student hypotheses (i.e., more stomata in the sun, fewer stomata with more CO_2); however, this was not always the case. Some plants showed a great deal of variation (e.g., some holly trees) that was unexplainable by treatment. Regardless, we noticed that as the students were collecting their data they were raising additional issues and posing hypotheses about characteristics of leaves, such as color and size, that may also have been affected by the environmental variable of interest. Clearly, their minds were working in unexpected directions and attempting to construct new relationships outside of the bounds of the inquiry. Often these insights and extensions generated lively discussions among students during the question/answer portion of the Stomata Results Symposium.

Student comments on this multi-week inquiry were generally very favorable. Many commented that they liked designing their own experiment and presenting their results to their classmates. Negative comments most commonly reflected student frustration with using the spreadsheet/statistical package available. During the second and third years we used improved instructional materials and tutorials, and students had been exposed to the software in new labs in their fall freshman lab course, so we received fewer of these comments.

Our impressions of the effects of this activity on *our* teaching are also positive. Considerable effort is required to resist giving too much information to the students during the project formulation stage and to simply "let go." However, this effort was returned when we witnessed the "eureka" of understanding that students attained typically when they saw the first graph depicting the results of their data.

The transfer of ownership of the inquiry from us to the students culminated in the Stomata Symposium during which we acted only as moderators of often lively student discussion. Importantly, many students who otherwise would be silent classroom occupants were fully engaged with their peers in this format. We get a great deal of satisfaction when we witness this kind of transformation.

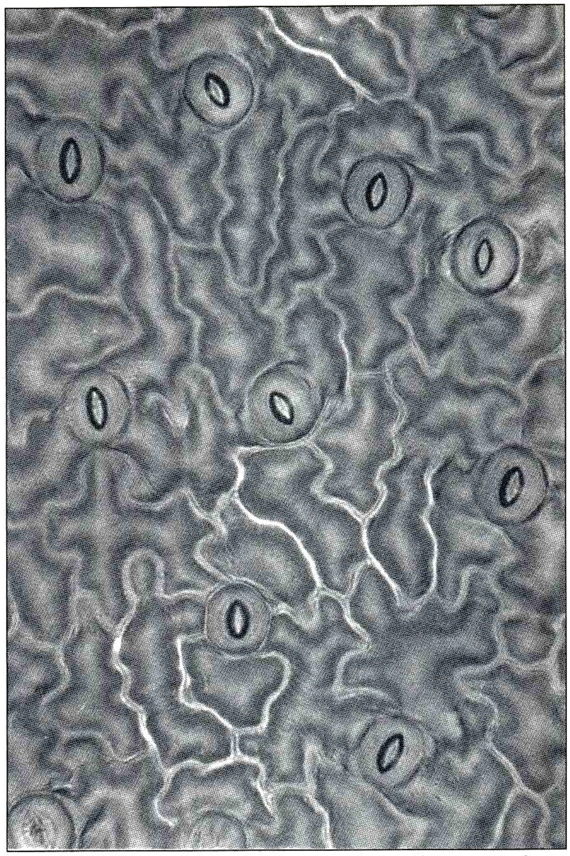

Figure 1: Stomata Impression. Photo showing what students would see in their nail polish stomata impression under a light microscope at 400x.

CONCLUDING COMMENTS

In closing, we wish to raise three points. First, it is important to realize that this activity requires two-and-a-half lab periods. We consider that affording students the opportunity to engage in all of the steps to a scientific investigation, i.e., "teaching science by doing science," is of paramount importance to freshmen and justifies this level of lab time allocation. These students get few other opportunities to develop and wield skills in experimental design, statistical analysis, and hypothesis testing in any content area of their curriculum.

In addition, plants are very distant organisms to most freshmen, and the intensity of this project may enhance their comprehension of the evolutionary principles of plant leaf design more so than conventional lecture formats alone. We gladly devote this seemingly large portion of the laboratory time available during the semester to achieve these objectives.

Second, dozens of recent reports on laboratories based on open-ended

Appendix 1: Detailed Procedure for Obtaining Stomata Impressions.

Obtain the leaf upon which you wish to census stomata.

On the side you wish to census stomata (typically the leaf underside) paint a rather thick swath of clear nail polish.

After the nail polish has dried (several minutes), obtain a square of VERY CLEAR tape (such as package sealing tape, but do NOT use scotch tape). Stick your tape piece to the area that contains the dried nail polish swath.

GENTLY, peel your nail polish swath from the leaf completely. You will see a cloudy impression of the leaf surface now attached to your tape piece. This is your "leaf impression." Make only one impression per leaf.

Tape your leaf impression to a VERY CLEAN slide and use scissors to cut off the excess.

Label the slide to indicate the treatment group name (e.g., leaf from sun) and other info (e.g., leaf #3) directly on the slide. Color coding slides is a very good idea.

Focus your leaf impression under at least 400x power and observe the stomata (see Figure 1).

Search around on your impression to find an area that subjectively appears to have a high density of stomata. That is, move the slide around until the field of view is away from the edge of the impression and so that there are no dirt blobs, no thumbprints, no damaged areas, and no big leaf vein impressions in view.

Count all stomata you see and record the number.

Repeat the previous two steps three times. The highest number of the three will be your datum from this impression. One datum per slide.

Repeat all steps above for at least eight different leaf impressions in each treatment group.

Students use a stage micrometer to convert their data from units of "stomata number per field of view at 400x" to units of "stomata per mm^2."

Note: Students must design their own data sheets on which to record stomata counts and they are informed that they will be specifically assessed in part on how well they accomplish the tasks of data acquisition and archiving.

student investigations in *JCST* and other science education journals provide important and creative directions for our collective teaching efforts. We must all try to work with our colleagues to explore these directions and make substantive changes *along course sequences* within major programs of study and not just within single courses. Educational meta-issues such as diversity, retention, and empowerment to think scientifically depend less on a student's experiences in a single course as upon his or her trajectory through the curriculum.

Third, we feel that an important goal of curricular innovation should also address the retention issue of teachers in pre-college settings and the valuation issue of teaching at the undergraduate level. As we mentioned above, we like teaching this kind of activity, and this attitude is often infectious with our students—and vice versa. At least for us, this affirmation instills a sense of renewal in our other academic pursuits.

Numerous authors have noted that open-ended inquiries give students a sense of ownership of their learning and that this contributes to their development as life-long learners. We wish to add that teaching using these types of activities promotes the same goal in teachers as well. ❑

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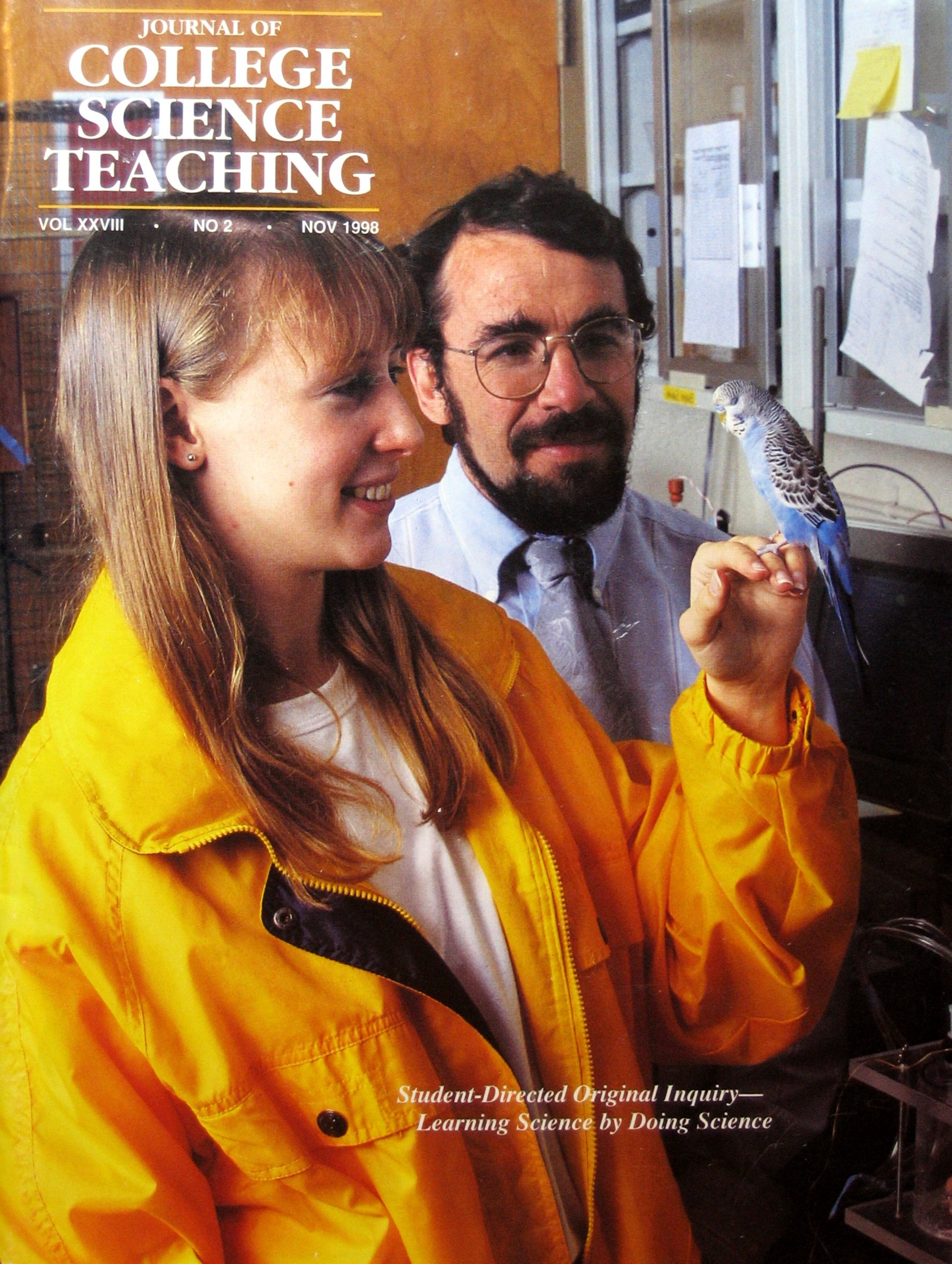

*Student-Directed Original Inquiry—
Learning Science by Doing Science*