INQUIRY & INVESTIGATION

Spiders by Night: An Outdoor Investigation Integrating *Next Generation Science Standards*

NABT BioClub RECOMMENDATION

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ABSTRACT

Field investigations represent an excellent opportunity to integrate the Next Generation Science Standards to complement and enhance both classroom and laboratory instruction. This inquiry-based exercise is designed to introduce students to the basic anatomy, ecology, and natural history of a common backyard denizen, the wolf spider (Lycosidae). *Students are charged with developing one or more testable hypotheses regarding wolf spiders in their own backyards. Wolf spiders are an ideal subject for field investigation because their secondary eyes possess a highly reflective layer called the* tapetum lucidum. *At night, this layer produces an unmistakable "eyeshine" when viewed with the beam of a flashlight. Playing the role of students, we tested the hypothesis that wolf spiders should occur at higher density in an undeveloped field than in a typical backyard. To test this, we utilized random quadrat sampling in both habitats using flashlights to detect nocturnal eyeshine. Students obtaining similar results would likely have concluded that wolf spiders were more abundant in natural habitats.*

Key Words: *Field study; nighttime survey;* tapetum lucidum*; quadrat sampling; wolf spiders; NGSS.*

○ Introduction

Scientific inquiry is an experience that should go beyond the books and benches of the traditional classroom and laboratory setting. The goals of the *Next Generation Science Standards* (NGSS; NGSS Lead States, 2013) are to demonstrate scientific proficiency through the integration of core scientific concepts, themes, and interdisciplinary practices. Ideally, implementing these standards would involve challenging students with open-ended questions while supporting their efforts to design and conduct independent investigations to explain natural phenomena (National Research Council, 2015). One often-overlooked strategy to spark student curiosity and encourage a growth mindset involves stepping outside of the typical classroom. Field research can be accomplished in even modest outdoor settings using basic tools (Hodgson et al., 2016) and established scientific practices including the development and testing of original hypotheses (Brownell & Kloser, 2015). Scientists who work with organisms common to many areas of the world (e.g., some types of plants, birds, or insects) see how useful these subjects can become for almost any type of project. Such investigations offer the flexibility for students to ask a multitude of questions as they document patterns ranging from species diversity and habitat use to the possible domination of experimental plots by one sex, species, or age group (Alahuta et al., 2014). Students who engage in projects such as these enhance their critical thinking skills and are much more likely to outperform their peers with a fixed mindset (Dweck, 2006).

> *Student-driven field research represents an ideal way to pique students' natural curiosity and integrate NGSS on a shoestring budget.*

Recognizing the importance of such investigations in STEM education, the Pacific Education Institute (2015) outlined a series of recommendations and strategies to encourage educators to incorporate field studies that help integrate NGSS to enhance student learning, presenting a variety of descriptive and comparative methodologies for field investigations that are commonly used by professionals in the environmental and natural resource sectors. For this inquiry-based exercise, we followed their recommendations and encourage educators to consider implementing simple field studies centered around the spider family Lycosidae. With over 240 species spread across 20 genera, wolf spiders are common throughout North America (Ubick et al., 2005; Bardier, 2015). Most species within this family are ground dwellers and hunt their prey at night (Ubick et al., 2005). Wolf spiders possess eight irregular eyes arranged in three rows. The two largest eyes, located in the middle row, possess a reflective membrane called the *tapetum lucidum* (Barth, 2002; Benson & Suter, 2013), which redirects scattered light

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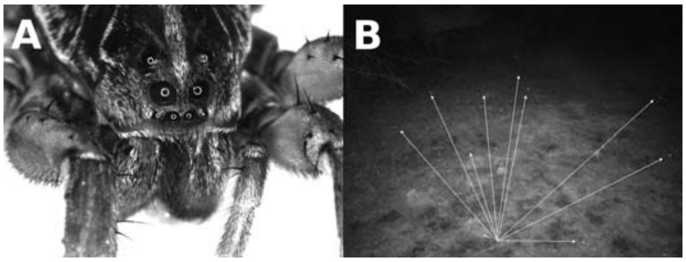

Figure 1. (**A**) The largest eyes in the center row possess the reflective *tapetum lucidum*. The white rings in the eyes are from the microscope light source (photo credit: J. Y. S. Hodgson). (**B**) The eyeshine of multiple spiders when illuminated by the flash of a camera (photo credit: A. Collier).

through the retina to increase visual acuity (Figure 1A). Using a flashlight to make the *tapetum lucidum* of the spider eyes shine (Figure 1B) can make it easy to locate and potentially capture these spiders at night (Bardier et al., 2015).

○ Independent Inquiry Promotes Novel Research

In an effort to promote independent lines of inquiry, we tailored this exercise for first- or second-year undergraduates or upper K–12 grade levels. This approach can easily be modified by providing more guided levels of instruction to better suit other grades as needed. As presented, it can serve as a modular exercise to supplement any biology-based course that introduces topics ranging from species diversity to ecology or evolution. We suggest that students be presented with a broad list of anatomical, ecological, and taxonomic questions regarding the family Lycosidae, similar to those presented in Box 1. Many of these questions are designed to be open-ended, providing students with only a framework as they begin their research.

Box 1. Suggested review questions.

- Spiders are classified as arthropods. What characteristics do all arthropods share?
- How do spiders differ anatomically from insects?
- Do spiders possess mandibles (jaws)? If not, how do they hold their food?
- How do male and female wolf spiders differ anatomically from one another? Could you identify the sex of an adult spider?
- Do most species of wolf spiders spin webs? If so, describe the webs they produce. If not, explain how they obtain their food.
- What is the *tapetum lucidum* and why do wolf spiders possess this structure? How is this structure beneficial to the spiders? What would happen at night if this structure was illuminated by the beam of a flashlight? What other animals possess a *tapetum lucidum*?
- Wolf spiders belong to the family Lycosidae. How would you describe most members of this particular family? Are they common in your area? In what types of habitats would you likely find them? What do they eat?
- What types of wolf spider species would you expect to find in your particular geographic area of the country?
- For the two spiders you named above, provide a description of the habitats in which they might be found. Describe their overall niche within these particular environments.
- How many eyes do wolf spiders possess? Briefly describe their basic anatomical position and overall function.
- Briefly describe (size, appearance, habitat use, foraging strategy, etc.) two commonly encountered species of wolf spiders in your area that potentially interact in nature as predator and prey.
- What is the largest species of wolf spider in North America? Does this species occur in your geographic area?
- What types of questions/hypotheses could you attempt to answer by examining populations of wolf spiders in your own backyard? How might you design an experiment to answer your question? Consider the materials and supplies you might require to perform such a project. What kinds of data might you collect and how would you assess and interpret your findings?

As students undertake this review, they naturally begin to connect many crosscutting concepts identified by the NGSS. These include learning about spiders' forms and functions, their niches within the environment, and the overall complexity and patterns of the natural systems in which they live. Ideally, students would have access to computers or tablets during a preliminary laboratory or lecture session. Box 2 provides a short list of some of the more informative online resources we referenced as part of our own review. Students could utilize a mix of these and other useful websites they uncover during their search, combined with any additional course-related resources to which they may have access. Students should be charged with developing one or more testable hypotheses regarding wolf spiders that can be examined in their own backyards or in other readily accessible habitats such as the school grounds or a nearby park. Wolf spiders are active year-round, although they may be more difficult to find during the coldest months of the year. With the help of their instructor, students will refine and narrow their objectives and begin to develop a sampling protocol to test their particular hypothesis.

Box 2. Information regarding wolf spiders and various field sampling techniques is readily accessible through these and other online resources.

Arachnids & Wolf Spiders

https://bugguide.net (identification and information about arthropods)
https://www.insectsofiowa.com (identification and preservation of arthropods)
https://www.insectidentification.org/spiders.asp (identification and home range of arthropods)

http://www.uky.edu/Ag/CritterFiles/casefile/spiders/spiderfile.htm (spiders of Kentucky)
https://en.wikipedia.org/wiki/Spider (general background information)
https://spiderid.com (identification of spiders using pictures)
http://www.bio.brandeis.edu/fieldbio/arachnids_cohen_weiner/Intro.html (arachnid identification)

Field Sampling Techniques
https://www.hawaii.edu/gk-12/opihi/classroom/measuring.pdf
https://mathbench.umd.edu/modules/env-science_sampling/page09.htm
https://www.webpages.uidaho.edu/veg_measure/Modules/Lessons/Module%205(Density)/5_2_Plot-based_Techniques.htm

To better illustrate how this exercise may unfold in your class, we played the role of students and proposed our own hypothesis to investigate as part of a pilot study. Here, we present our experimental design and describe the methods utilized to collect data in the field. We used simple descriptive statistics to analyze and interpret our findings, and multiple evaluative strategies to assess the overall merits of our particular project. Recognizing the limitations of our pilot study, we recruited a small number of K–12 students to replicate our collection efforts and identify problems in our sampling design. In addition, we describe a number of other potential field projects that other students might choose to explore as part of their own investigation. The student learning outcomes (SLOs) for this inquiry-based exercise were adapted from the Pacific Education Institute (2015):

- SLO 1: Develop information literacy; activities: search printed literature and Internet sources to summarize a broad topic of interest.
- SLO 2: Formulate investigative questions; activities: ask questions about abundance and differences between groups, conditions, or habitats.
- SLO 3: Demonstrate systems-level thinking; activities: identify habitat types and associated structural and organismal differences between them.
- SLO 4: Identify variables of interest; activities: choose variables to be measured in at least two different locations or measured together to test for relationships.
- SLO 5: Formulate a hypothesis and collect data; activities: propose a hypothesis based on identified variables, design a sampling protocol, and collect data.
- SLO 6: Analyze and interpret data; activities: plot and statistically analyze data to elucidate patterns for comparison to other systems.

○ Pilot Study

Does Wolf Spider Density Differ between Habitats?

After addressing the review questions (Box 1), we chose to investigate whether the density of wolf spiders varied between different habitats. We hypothesized that they would be more prevalent in an undeveloped field than in a typical urban backyard. We utilized a comparative field investigation (Pacific Education Institute, 2015) to study the differences. The following protocol was designed to sample these habitats, and our data are presented and analyzed in much the same way that students would use with their own projects.

Methodology & Data Analysis

We used quadrat sampling to test our hypothesis regarding spider density in two distinct habitats. This sampling technique is commonly used to determine a species' presence or absence and to provide estimates of overall abundance and density (Jaeger & Inger, 1994). The protocol first involved dividing the habitat of interest into a rectangular grid of equal-sized units (Jaeger & Inger, 1994). For our study, we used a meter wheel and surveyor flags to divide a 0.10 hectare (0.25 acre) backyard habitat into four square grids of equal size (length 30 m × width 30 m = 900 m^2). This habitat was largely comprised of a uniform turf of centipede grass (*Eremochloa ophiuroides*). By comparison, the undeveloped field was ~0.41 hectares (1.0 acre) characterized by sandy soil with a mix of bahia grass (*Paspalum notatum*), southern wiregrass (*Aristida beyrichiana*), and patches of dogfennel (*Eupatorium capillifolium*) among other invasive species. For consistency, we laid out an identical grid consisting of four squares of equal size (900 m^2) centered in the interior of the larger field (see protocol references in Box 2). We constructed a circular 1 m^2 quadrat using ¾ inch plastic tubing purchased at a home improvement center for under $15. Students could also use a standard hula hoop as a quadrat but would first need to calculate the internal area as follows: area = $\pi(d/2)^2$ = __ m^2.

Prior to sampling, we recorded the time and temperature and provided a basic description for each habitat in our field journal. We sampled at night, entering each square of our rectangular grid at a randomly chosen compass direction and walked 10 paces before dropping the quadrat. Flashlights were held at the temple with the beam pointed downward at a 45° angle to maximize eyeshine detected. The number of spiders visible from our original position was recorded, and we proceeded to encircle the hoop to identify any others that faced a different direction. Although the spiders commonly shifted positions or partially retreated into their burrows, few moved more than short distances when fully illuminated. Upon completion, we retrieved the hoop and walked 10 paces in a randomly chosen compass direction before dropping the quadrat and repeating the process. For both field and backyard habitats, we counted the number of spiders in 10 randomly selected quadrats for each square of the grid. We sampled on only one occasion, to highlight the extent of data that could be collected by students in just one evening (total data collection time: ~30 minutes per habitat). For this field protocol, each quadrat represents an independent sample, and statistical inferences can be drawn from the data, assuming that the sample size is sufficiently large (Jaeger & Inger, 1994). Although our time in the field was limited, we still collected data from a total of 40 quadrats in each habitat and were able to make statistical comparisons. Using the Analysis ToolPac feature available in Microsoft Excel, we compiled our data and tested the following null and alternative hypotheses using a two-sample *t*-test:

- H_o = The abundance of lycosid spiders does not differ between habitats.

- H_a = The abundance of lycosid spiders differs between habitats.

Wolf spiders encountered in the field can readily be identified to family. If students select a project requiring more detailed identification (to genus or species), the resources found in Box 2 can be utilized. Spiders can be collected safely using forceps and then both euthanized and stored in isopropyl alcohol for future identification. In our field sampling, we took photographs of spiders with an iPhone 6, using the flash setting to capture reflective eyeshine.

Results

We encountered an average (± SE) of 1.6 ± 0.160 wolf spiders per square meter and a total of 62 individuals in the backyard habitat. Conversely, we identified 101 individual wolf spiders, or an average of 2.5 ± 0.232 per square meter, in the undeveloped field (Figure 2A, B). The abundance of wolf spiders differed significantly between the two habitats (t = 3.15, P < 0.01). We were therefore able to reject our null hypothesis (H_o). The most common species we encountered was the Carolina wolf spider (*Hogna carolinensis*; Figure 3A). This is the largest wolf spider in North America, and their large eyes reflected intensely when illuminated (Ubick et al., 2005). The characteristic eyeshine was an effective technique for identification of *H. carolinensis*, even before the spider was approached. We also collected smaller individuals belonging to the genus *Pardosa* (Figure 3B) but did not identify these beyond that taxonomic level.

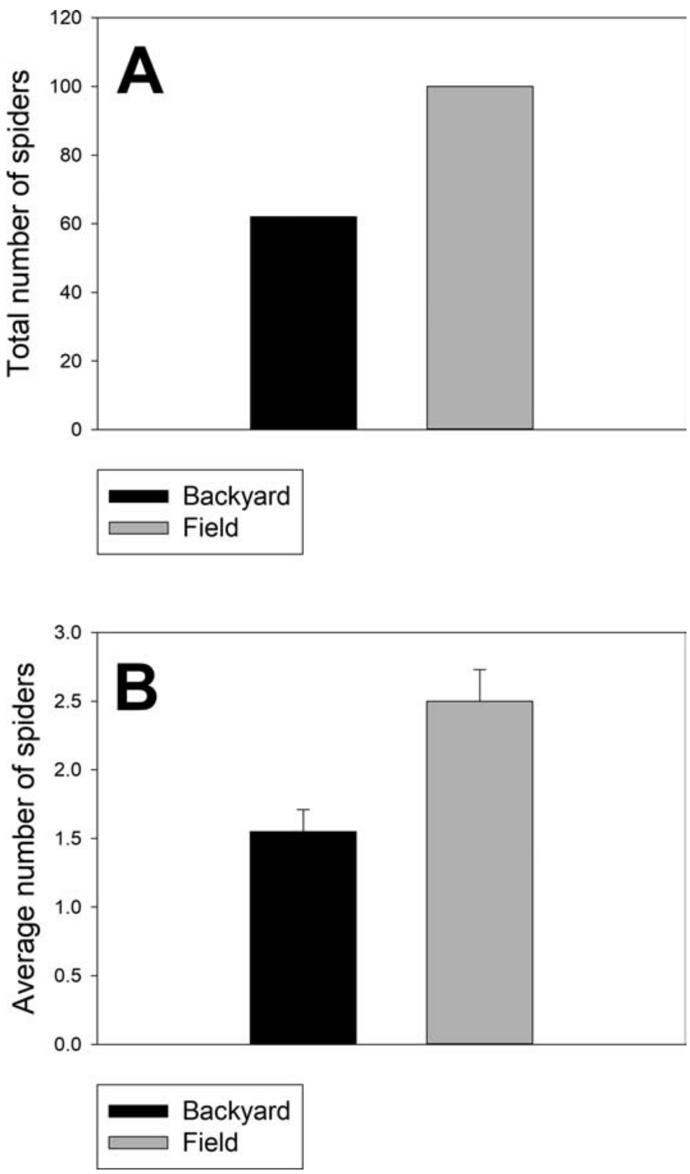

Figure 2. (**A**) Total number of spiders observed and (**B**) average number (± SE) of spiders per square meter in backyard and field habitats.

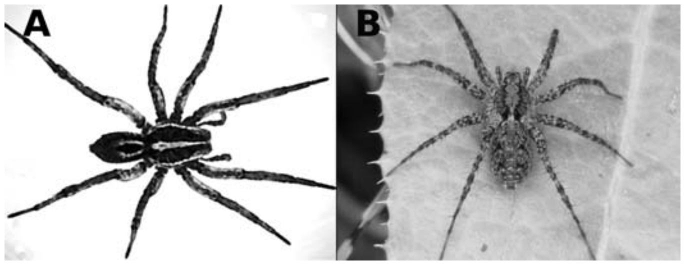

Figure 3. (**A**) *Hogna carolinensis* (photo credit: K. Craven). (**B**) *Pardosa* sp. (source: Wikimedia Commons; full citation is provided in the References).

○ Student Feedback & Troubleshooting

Data were collected for the pilot investigation during the summer, with two small groups of K–12 students, to test our hypothesis-driven activity and assess the methods. The first group consisted of three eighth-graders who utilized the same habitats we originally explored. The second group consisted of four fifth-graders who collected data from an entirely different backyard and field habitat. We met with both groups separately to discuss the project and asked each to tackle a subset of the review questions (Box 1). The students were also given the opportunity to use a flashlight in a dark closet to detect the eyeshine of a living spider we had collected and placed in a clear jar. After a brief introduction to our chosen sampling methodology, we provided both groups with a meter wheel and a handful of surveyor's flags and headed out to their chosen habitats before the sun went down. Students were observed as they mapped out their habitats in roughly the same relative shape and size. As sampling commenced, both groups did an admirable job of tossing their hoops in random compass directions and worked diligently with their flashlights to record the number of visible spiders within each quadrat. Students collected samples throughout each habitat; however, not enough replicate samples were collected to permit statistical analysis.

Feedback from the students who participated in this project was positive. All of the students involved in the testing appeared to thoroughly enjoy the process of throwing their hoops and counting spiders in the beam of the flashlight. Although they liked working with the meter wheel and flags, many failed to understand why these grids were important to the study, and most thought this step could have been eliminated. While we still encourage instructors to introduce this protocol, we acknowledge that it might present an unnecessary hurdle for certain students. Instructors who teach younger grade levels, or those supervising students who will need to commute longer distances to collect data, might choose to forgo this preliminary step. In the absence of establishing a primary grid, instructors are strongly encouraged to stress the importance of collecting random samples throughout each habitat. Based on the success of this method using K–12 students, we are confident it can be scaled appropriately to challenge college-level students as well.

○ What Might Your Students Investigate?

Students may pose any number of suitable questions and hypotheses regarding the family Lycosidae. To an instructor with limited knowledge of spider biology, this may sound potentially daunting. We stress, however, that none of the authors had previously worked with wolf spiders or arthropods in general. By acknowledging the use of this family as model organisms, we can explore a multitude of possibilities. For example, students could conduct a simple inventory to assess species diversity. Their sampling protocol may involve sampling spiders at night, as previously described, and later properly identifying each by genus. Several of the websites listed in Box 2 provide user-friendly resources for this purpose. Other students may instead compare the density of spiders found in different microhabitats within the same backyard or field using similar protocols we have adopted. They could easily determine whether spider density differs in proximity to a fence or driveway compared to a grassy area. Clearly, there are far more lines of potential inquiry and sampling methodologies that students may propose and implement than can be adequately summarized here.

○ Considerations about Equipment, Supervision & Expectations

Depending on the question and scope, students could require a combination of sampling methods to properly implement their study. Instructors should familiarize themselves with some of the more common field techniques that support a wide range of student-driven projects (summarized in Pacific Education Institute, 2015). Students can also consult various online resources that outline inexpensive yet effective protocols (see Box 2). While labor intensive, these techniques require relatively inexpensive supplies and materials. Our meter wheel, surveyor flags, and the materials for our homemade quadrat were purchased for under $40. These items are reusable and could easily be shared by the class members to minimize overall costs. Additionally, many universities and K–12 classrooms have modest budgets that can be used to purchase the basic sampling equipment outlined in this exercise. Instructors and students are encouraged to brainstorm and develop ways to use existing classroom equipment and/or more inexpensive alternatives as their gear if budgeting is an issue. Assuming that students have access to field-friendly clothes and basic supplies (rain apparel, boots, insect repellent, field journals, flashlights, etc.), they should be able to conduct their projects with little out-of-pocket expense.

This exercise involves nighttime sampling, which may be a concern. We feel that it is reasonable for university instructors, especially at residential campuses, to facilitate and/or lead this guided inquiry at night. Nighttime activities are commonplace for many university courses, particularly astronomy, but also for biology and geography. In a K–12 setting, we also feel it is reasonable for teachers to organize and supervise a twilight or after-dark activity on school grounds for students who may not have access to a yard. It is customary for schools to organize club meets, sporting events, and field trips for students after dark. All activities should be conducted within established institutional safety guidelines. Also, urban settings, such as a college campus or K–12 schoolyard, are highly effective environments for teaching basic tenets of habitat ecology and guided inquiry of ecological concepts using NGSS templates (Hodgson et al., 2016; also see Lundholm & Richardson, 2010). Therefore, students will be able to successfully satisfy the goals of this exercise, without compromise, in a variety of locations.

Overall, it is important to note that even the best-laid plans to collect field data are often met with unexpected hurdles. Many student groups will realize that their original sampling protocol or research focus was simply not practical. Although frustrating, this trial-and-error approach may help students better appreciate that the scientific method is anything but a simple linear process (Pacific Education Institute, 2015).

○ Assessment Strategies

For this exercise, SLOs 1–4 are front-loaded and provide the instructor with early feedback regarding student progress. Collectively, each group of students should demonstrate a baseline level of information literacy as they review the natural history of their study organism. Early success will be measured by their ability to formulate one or more investigative questions and demonstrate systems-level thinking. Students should submit a short (one- to two-page) research proposal that summarizes their particular question or hypothesis and clearly identifies the variable(s) that will be measured or observed. This will serve as an early evaluative tool to assess their overall progress and can be used to help them select the sampling protocol best suited for their project.

Throughout this process, students should keep a detailed field journal that tracks the development of their project and documents their sampling efforts in the field. The instructor will need to provide guidance and set clear standards regarding how these data entries are to be organized. With the appropriate rubric, these journals will serve as documents for future assessment.

In addition, we suggest that each group be required to present their findings in the form of a scientific poster or triptych during a future class or laboratory session. Individual students could also demonstrate their level of proficiency by explaining a different part of their group's project to the class. Students will describe their research methods, data collection, analysis, and interpretations as stressed in SLOs 5–6. At the advanced student level, a written research report summarizing the project provides a capstone experience. These options provide the instructor with flexibility to assign some mixture of potential assignments while still gaining a wealth of material to evaluate whether students have begun to develop deeper learning strategies. Instructors also have the option to develop pre- and post-assessment questions that could provide additional feedback to track the effectiveness of such projects as they pertain to long-term goals and expectations. We have included a representative rubric that matches the SLOs we outlined (Table 1).

Table 1. A sample rubric for grading this activity. Student learning outcomes (SLOs) are listed in the text. Grading scale: 16–18 points = A; 14–15 points = B; 12–13 points = C; 10–11 points = D; <10 points = F.

SLO	Did Not Meet SLO Goal: Fail (0 points each)	Met SLO Goal: Fair (1 point each)	Met SLO Goal: Good (2 points each)	Met SLO Goal: Excellent (3 points each)
1	No citations	1–2 citations	3–4 citations	5–6 citations
2	Did not ask a question	Asked a question about 1 variable	Asked a question about 2 variables	Asked a question about 3 or more variables
3	No identification	Identified habitat	Identified habitat and structural differences between habitats	Identified habitat, structural differences between habitats, and organismal differences between habitats
4	Did not select variables	Selected a variable in 1 location	Selected a variable in 2 or more locations and measured them together	Compared a variable collected together in 2 or more locations
5	Did not propose hypothesis	Proposed hypothesis	Proposed hypothesis and designed sampling protocol	Proposed hypothesis, designed sampling protocol, and collected data
6	Did not organize or compile data	Plotted data	Plotted and statistically analyzed data	Plotted, statistically analyzed, and compared data between ecosystems

○ Conclusions

We strongly support the recommendations outlined by the Pacific Education Institute (2015). Student-driven field research represents an ideal way to pique students' natural curiosity and integrate NGSS on a shoestring budget. Although students could study any number of organisms in the field, we recommend that they consider working with lycosid spiders. With their characteristic eyeshine, wolf spiders are easily located at night and students can use them in novel ways as a model organism to test any number of hypotheses. Our pilot study regarding the abundance of wolf spiders in differing habitats was easy to conduct, and the reflection from the eyes of dozens of spiders kept our adrenaline pumping and gave groups of K–12 students a hands-on activity to remember. As "students," we conducted our own initial review, posed a testable hypothesis, and collected and analyzed data using standard field sampling techniques. Collectively, these efforts mirror the overall goals for implementation of NGSS outlined by the National Research Council (2015). Projects such as these may help students acquire the mindset that they can become strong, independent learners and help propel them toward future success in STEM-related careers (Dweck, 2006).

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