Assessing the Importance of Chemosensory Cues in Web-Building Spiders: An Inquiry-Based Investigation for Secondary Education Students

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ABSTRACT

Spiders rely on chemosensory cues to help locate prey, evade predators, and select mates. This reliance on chemosensory detection makes spiders an ideal organism for inquiry-based projects that students can design and conduct on a shoestring budget. This investigation, designed for secondary education grades (6–12), encourages students to examine whether chemosensory cues influence web construction of harmless female spiders collected from backyard habitats. Students work in groups to develop hypotheses that they later test by introducing spiders into a simple T-maze containing some mix of chemosensory cues. To pilot this study, I designed two experiments to determine how female spiders construct their webs in response to chemosensory cues of potential prey or dangerous fire ants. The data from both projects were compiled and statistically analyzed using open-source software available online. In a classroom or laboratory setting, your students can work in groups to develop their own hypotheses, design and run their experiments, and statistically analyze their results using the same free software. Although simple in design, this activity provides students with an opportunity to develop novel lines of research inquiry and engage in the practice of science.

Key Words: inquiry-based learning; NGSS; chemosensory detection; spiders.

INTRODUCTION

Spiders possess chemosensory setae (hairs) on their walking legs and pedipalps that detect substrate (contact) and airborne chemicals in their environment (Nentwid, 2013; Ganske & Uhl, 2018). The pedipalps are small jointed structures located between the chelicerae (jaws) and walking legs that are also used in courtship displays and help males transfer sperm during mating (Wikipedia, 2021). The setae detect chemical cues of potential predators (Bell et al., 2006; Nentwid, 2013) and help spiders track prey and select mates (Kasumovic & Andrade, 2006; Nentwid, 2013; Giroti & Brescovit, 2019; Fischer, 2019; Persons & Rypstra, 2001). Spiders therefore represent an excellent model organism that may react in predictable ways when exposed to certain chemosensory cues. The spider-centric exercise described here incorporates many of the recommendations outlined by the Next Generation Science Standards (NGSS), in which students learn core ideas and crosscutting concepts through first-hand engagement in the practice of science (NGSS Lead States, 2013). With only a brief overview regarding the diverse manner in which spiders rely on chemical communication to survive, students can develop their own testable hypotheses regarding how these cues influence web construction of spiders inside a simple T-maze. Students will also learn to identify and better appreciate the natural history of many common backyard species. With instructor supervision, they can also collect their own spiders and can even work in groups to safely construct the T-maze chambers described in this activity.

CONSTRUCTION OF T-Mazes

T-mazes can be easily, and relatively inexpensively, constructed using PVC pipe and other supplies that can be purchased from most home improvement retailers (Table 1). To pilot this study, I constructed five maze chambers that would be sufficient for a typically sized classroom or lab (Figure 1). The arms of each maze were 110 mm in length and were designed to be separated from the middle PVC T-fitting so that different chemosensory cues could be introduced into only one arm of the chamber. Additional details regarding construction and assembly are provided in Appendix A (available with the online version of this article). Spiders are subsequently introduced into the pitch-black maze and must rely on a mix of tactile and chemosensory cues to help construct their web.

PILOT STUDY & HYPOTHESES

I piloted this project in summer 2020 during the height of the Covid-19 pandemic and was unable to recruit student participation due to CDC guidelines regarding social distancing. Nevertheless, I played the role of a student and posed two testable questions for this project: Do spiders preferentially construct their webs in areas rich with chemosensory cues of potential prey? Do spiders avoid constructing webs in areas containing chemosensory cues...
of dangerous fire ants? Because I would be collecting discrete, or nominal, variables regarding the presence or absence of a spider in a particular arm of the T-maze, my null and alternative hypotheses for both lines of inquiry were identical.

\[ H_0 = \text{Spiders/webs would be randomly distributed (50:50) between maze arms.} \]

\[ H_a = \text{Spiders/webs would not be randomly distributed between maze arms.} \]

○ Spider Collection

Most spiders are harmless to humans, and bites from backyard species are relatively uncommon. Although all spiders produce venom, most possess small fangs that cannot puncture the skin. Nevertheless, care should be taken when identifying a particular backyard web builder for this activity. Instructors can determine if potentially venomous species are located in their particular state using online resources such as www.insectidentification.org. Although spiders can be safely collected and relocated using simple instruments to avoid direct contact, students who are sensitive or allergic to insect bites and bee stings should exercise particular caution. Students are encouraged to wear a pair of garden gloves when collecting and handling spiders.

Prior to the start of the experiment, I surveyed my own backyard and surrounding neighborhood in search of suitable web-builders. I encountered the majority of webs along fence lines and under the roof of a backyard shed. Apart from steering clear of widow spiders which are common in my area, I took pictures of potential spiders, which I later identified with a simple online search (www.insectidentification.org).

I chose to work with the orchard orbweaver spider (\textit{Leucauge venusta}) which is one of the most common species in the eastern United States (Hall, 2019). Females of this small (5.5–7.5 mm) and colorful spider are easily located as they tend to hang upside down (Figure 2) at the center of their horizontally constructed web. Females have a habit of dropping from the center hub of their web on a silk line when disturbed, and it was quite easy to collect them

![Figure 1. Completed T-maze chambers with arrows highlighting location where spiders were later introduced.](image-url)

![Figure 2. Orchard orbweaver (\textit{Leucauge venusta}) female on a web prior to collection (photo credit: T. Collier).](image-url)
in a small container. I later used a mix of craft (popsicle) sticks, a metal lab spatula and the occasional paper cup to transfer each spider to a particular T-maze.

- **Testing the Hypotheses**

To test the first hypothesis, I collected small grasshoppers as “prey” from local habitats and isolated three individuals in one arm of each T-maze overnight. At this time, cap fittings were placed on both ends of the maze arm to prevent prey escape. The other arm of the maze contained no chemosensory cues and served as the control. For the second hypothesis, I collected sandy soil from the edge of fire-ant mounds and allowed it to dry completely in the sun. I later added a half cup of this dry (ant-free) sand into one maze arm of each chamber. In the second arm I placed a half cup of dried play sand that contained no ant chemosensory cues. Cap fittings were placed on either end of each sand-filled maze arm. For each study, the chemosensory cues were left in place overnight (18 hours), the insect prey were later released unharmed, and the sand was drained from each tube. At the start of each trial, the treatment and control maze arms were reassembled (see Appendix A), and spiders were introduced into the middle PVC T-fitting (Figure 1, arrow). The rubber stopper was then inserted to seal the opening. The spiders were left undisturbed overnight to construct their webs in either arm of the T-maze. The following morning I removed the stopper from each maze and inserted the inspection mirror at an angle to identify the location of the spider and her web. At the end of each trial after the spiders were removed, the maze arms and T-fittings were disassembled and cleaned with alcohol. Spiders were temporarily housed in separate small containers between trials and were later released unharmed. For each line of inquiry (prey and fire-ant cues), I conducted 15 replicates and ran each spider in two separate maze chambers to reduce the total number needed for this study (n = 15). Without exception, the spiders connected long silk strands at both ends of the maze armature, but the web was more densely concentrated in one arm of the chamber where the female resided. The data collected for both projects are shown in Figures 3A and 3B.

Nominal variables, such as the presence/absence of data from this study, are often statistically compared using a chi-square test. However, because of the small sample size there was a greater risk of committing a Type I error (incorrectly rejecting the null hypothesis). Because of this, data were compared using separate two-tailed exact binomial tests of goodness of fit using an online statistical spreadsheet (McDonald, 2014; http://www.biostathandbook.com/exactbin.xls). Unlike most other statistical tests, an exact test, such as the one described here, does not calculate a test statistic that is used to estimate the probability of obtaining the observed data. The exact test instead directly calculates the probability based on the null hypothesis (McDonald, 2014). This test is commonly used when considering one nominal variable with a relatively small sample size (McDonald, 2014). For this test, the null hypothesis is only rejected if the calculated probability (p value) is less than 0.05. Although web construction was more common in maze arms associated with prey cues, these data were not statistically significant (p = 0.607). Likewise, there was no statistical difference in the placement of web construction between the control maze arms and those containing cues of fire ants (p = 0.118).

- **Conclusion**

Due to Covid-19 restrictions, I was unable to pilot this activity with the help of students and unfortunately cannot provide direct feedback regarding large-scale implementation and assessment in the classroom. Nevertheless, I remain convinced that this project is suitable for grades 6–12. With proper supervision, your students can safely construct their own maze chambers (see Appendix A), use online resources to confidently identify harmless species, and later collect them from their own backyards or local neighborhood. Although anecdotal, my own kids (ages 10–16) helped me construct the chambers used in this pilot study and periodically collected spiders and data with little difficulty. Overall, the experimental design is quite simple and the collection of data and statistical analysis should not be an impediment for students of this age group.

Your students should not be limited to the hypotheses tested in this pilot study. Using the same maze chamber design, they could develop any number of trials that could instead expose female spiders to chemosensory cues of potential predators, competitors, or even possible mates. The results from the pilot study nevertheless illustrate an important point that your students may also encounter when collecting and analyzing biological data. Even when observable patterns are documented, those results will not necessarily prove to be statistically significant. Students may also begin to appreciate some of the limitations and flaws inherent in their original study.
design only after they begin collecting data. For example, I made a poor choice when selecting grasshopper “prey.” Common prey of the orchard orbweaver actually include much smaller species of flies and mosquitoes belonging to the order Diptera (Hall, 2019). Recognizing the limitations of your study design and acknowledging early missteps is a necessary part of scientific inquiry (Brownell & Kloser, 2015). Although relatively simple in design, students can posit any number of testable hypotheses using the maze chambers described here. Inquiry-based investigations such as the one described for this activity represent an important opportunity for students to engage in the practice of science.

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