Using a Combined Approach of Guided Inquiry & Direct Instruction to Explore How Physiology Affects Behavior

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Abstract

Hands-on activities with live organisms allow students to actively explore scientific investigation. Here, I present activities that combine guided inquiry with direct instruction and relate how nutrition affects the physiology and behavior of the common housefly. These experiments encourage student involvement in the formulation of experimental design, promoting engagement in the learning process. These activities are suitable for both postsecondary education and high school classroom settings and highlight National Science Education Standards, particularly by promoting inquiry-based learning and communicating science explanations.

Key Words: *Behavior; physiology; Musca domestica; inquiry-based learning; scientific process; experimental design; IPM; nutrition.*

Undergraduate laboratories provide a unique environment for students to apply the principles of scientific process through inquiry-based learning, though these opportunities are often overlooked (Hughes & Ellefson, 2013). Students need exposure to authentic research as science standards increasingly focus on question- or problem-driven, inquiry-based learning to engage students in the processes of research and scientific discovery (National Research Council, 1996; Boyer Commission, 1998). However, many laboratory classes still perform standardized experiments, highly predictable in experimental design and outcomes, to teach basic scientific skills (Hughes & Ellefson, 2013). These confirmatory experiments do not allow for independent and critical thinking by the student. Complicating instruction is the inadequate exposure students receive to scientific inquiry, which limits their skills in developing sound research questions and methodology. Guided-inquiry experiments can enhance the learning experience of students and expose them to scientific principles (Marshall & Dorward, 2000); when instructors retain sufficient control over the process, they can help students develop scientific skills through the activities (Wang & Lin, 2008; Ku et al., 2014).

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The inquiry-based activities described here are original applied-research experiments (Aditomo et al., 2013) that use a combination of guided inquiry and direct instruction to develop scientific skill sets. Using house flies (*Musca domestica* L.), students are exposed to the scientific inquiry by investigating how housefly feeding preferences are related to the cause-and-effect relationship of physiology and behavior, and how these principles influence fly control. Students begin by developing a hypothesis and, with guidance, methods to test the hypothesis; then they use critical thinking and cognitive skills to discuss results. Students critically analyze the basic physiological drive for foraging and synthesize results, apply biological principles and discuss animal behavior and nutrition, and consequently develop skills to apply those principles to other science-related activities. Additionally, these experiments require minimal materials and can be conducted in a short timeframe.

Insects are ideal animals to use for biological studies in the classroom (Matthews et al., 1997). They are easy to obtain and rear, commonly model other organisms or biological processes, and often produce quick results. Houseflies, in particular, are an easily recognized and familiar insect. These flies are considered synanthropic, or found in close association with humans and their environment. Though houseflies are often considered only pests, they offer many opportunities for better understanding insect physiology and behavior and their influence on control.

These experiments were conducted in an undergraduate introductory entomology laboratory for majors and nonmajors with the aim of engaging students in the research process. These activities could easily be incorporated into science frameworks under the *National Science Education Standards* and can be adapted for grades 9–12 or postsecondary education science classrooms.

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○ Background

Adult houseflies have a wide range of suitable food sources, but food searching behavior is likely associated directly with nutritional needs based on ovarian development and energy requirements. Houseflies require carbohydrates and protein at various stages of their adult life (Goodman et al., 1968). Carbohydrate energy sources like sucrose are critical to the survival of adult flies. When deprived of carbohydrates, sugar feeding has been cited as taking precedence over other nutritional needs (Greenberg, 1960). Conversely, houseflies are anautogenous, which means that they also need protein as adults for egg maturation. Ovarian development occurs similarly between newly emerged adults fed sucrose and protein for about 2 days; however, in the absence of protein, further ovarian development ceases at that point (Goodman et al., 1968). The following experiments test the foraging behavior of gravid or nongravid female flies or of newly emerged flies fed protein or carbohydrates exclusively.

○ Student Preparation

For the experiment to be effective in stimulating critical thinking and analytical skills, students must be sufficiently prepared with the relevant background information and skill set to conduct and interpret the experiment. Though this was developed for a university midlevel science course, many non–science majors are enrolled, and the material is suitable for high school and postsecondary classroom settings. Therefore, a basic review of scientific process and experimental design was required. This review was provided as an information packet and not presented as lecture material. Through this packet, students were also introduced to the basic elements and requirements of a scientific paper.

Students were required to read relevant refereed journal articles (e.g., Goodman et al., 1968; Greenberg, 1959) that provided not only a model for a journal article, but also background information that could assist with formulating a hypothesis and generating ideas for methodology and evaluation of the data.

Students were engaged in a class discussion about the nutritional needs of houseflies, including how protein and carbohydrates affect physiology and behavior. The influence of physiological state (e.g., age or ovarian development) on flies' nutritional needs was discussed, as well as how variation among proteins and carbohydrates (e.g., in water content, color, amount, or viscosity) might change behavior or feeding time. Students were asked to generate a list of possible sources of carbohydrates and protein that might be attractive to houseflies. They were then given materials for the experiment, and the instructor engaged the students in a discussion of how these materials could be used to test the feeding behavior of flies – including how to provide the substrate, timing of the experiment, age to test flies, and so on – thus effectively leading students through the formulation of a basic experimental design.

Students were assigned to groups of no less than three and up to six, depending on the size of the class. Each group chose one protein and one carbohydrate source from the available options (see below). Groups were then given a worksheet to discuss the following points related to the experiment:

- Description of experimental dependent and independent variables
- Number of replicates to conduct
- Description of the control
- Specific hypothesis based on substrate choice
- Statistical analysis to be used
- Predicted table or figure to be constructed from collected data

The experiments described below are two examples of the types of experimentation that can be conducted with these materials. Depending on the level of student input and creativity, these experiments can be adapted for more independent methodology to explore substrate preferences in choice-tests at various fly ages or other questions generated by students. Other modifications to allow for more student input include substrates chosen by students and provided by the instructor and dishes of different dimensions provided as testing arenas.

○ Initial Experiment Preparation

Materials & Methods to Prepare for the Experiment

- Housefly puparia
- 50-mm plastic Petri dishes or ≤8-oz plastic containers
- Propane blow torch (if using Petri dishes)
- 1-cm-diameter metal rod (e.g., rebar) (if using Petri dishes)
- Safety glasses (if using Petri dishes)
- Small scissors (if using plastic containers)
- Cotton
- Plastic bowls
- Soft forceps
- Powdered egg yolk
- Powdered milk
- Table sugar (sucrose)
- Screened cage (30 × 30 × 30 cm or larger)
- Black cloth
- Rubber band

Plastic Petri dishes are reusable and are the preferred method for observing housefly behavior, but it is possible to use ≤8-oz plastic containers if the plastic is clear. Plastic containers may be reusable, depending on the quality. If using Petri dishes, the instructor should prepare these materials by igniting the blowtorch while wearing appropriate personal protection. The metal rod should be heated and placed on the lid of the Petri dish, burning a single hole through the top of the dish, close to the outer rim of the lid (Figure 1). The plastic containers are prepared similarly, but instead of burning a hole, small scissors can be used to cut a hole about half an inch to one inch in diameter.

Housefly puparia are readily available for purchase through an educational biological supply company. Six flies will be needed per student, and it is recommended that at least twice as many flies are purchased than are needed, to account for unexpected mortality. Adult flies will emerge in 3–4 days, or can be refrigerated if delayed emergence is needed. Approximately 1000–1500 puparia should be placed in a 250-mL cup and put in each of two screened cages with good ventilation (Figure 2), or four cages if both experiments

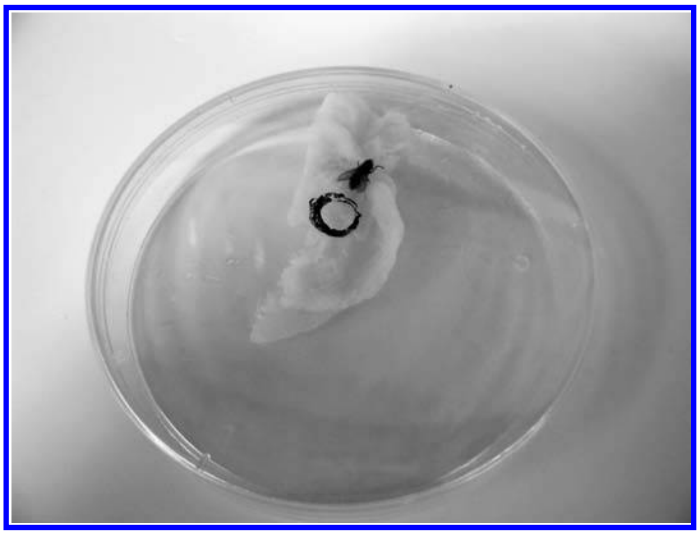

Figure 1. Petri dish used during both experiments, with the rolled-up gauze and carbohydrate substrate (sports drink) placed perpendicular to the edge of the Petri dish.

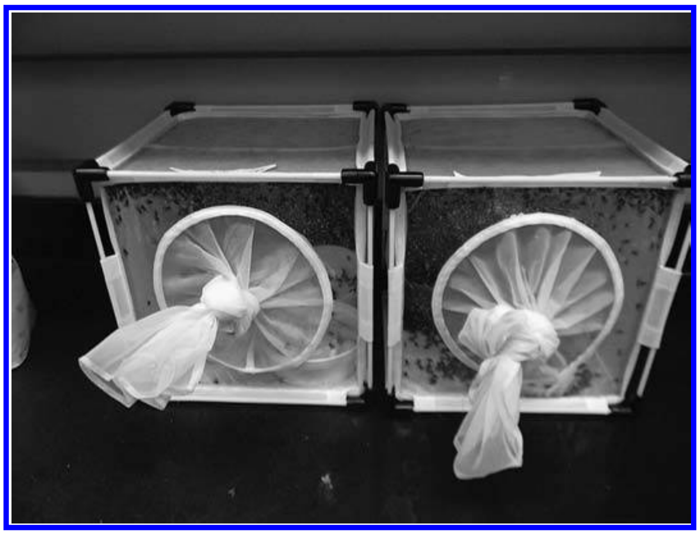

Figure 2. Screened cages (30 × 30 × 30) for house flies.

are being performed. Collapsible screened cages (12 inches) can be purchased from Bioquip (Rancho Dominguez, CA) for less than $11.00 per cage. Cages can also be made with a 1 × 1 foot frame and window screening. Water should be available to flies at all times. Provide flies with 250 mL of water and add foam chips or a folded piece of paper towel to prevent drowning.

The day before the experiment, flies should be separated by sex and females placed in a separate ventilated container with access to water only, to facilitate active foraging during the experiment. Females can be recognized by the relative space between the eyes. In males the eyes almost touch, whereas female eyes are distinctly more separate (Figure 3). Flies can be placed in a container on ice or in a freezer for 5–10 minutes to decrease activity. The day of the experiment, flies can be slowed again with the same technique as above and individually placed in dishes. Flies should be given ≥10 minutes to acclimate to the dishes prior to testing.

Little or no experience in rearing insects is needed to prepare for these experiments. Adult flies will need to be fed only once,

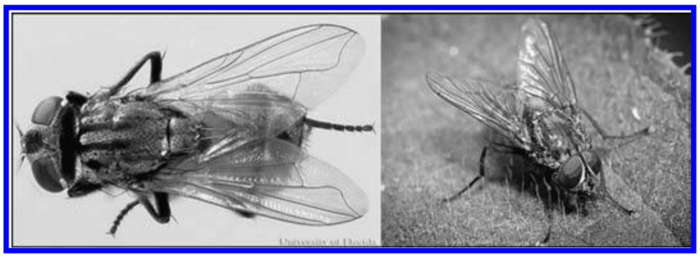

Figure 3. Comparison of eye distance in female (left) and male (right) houseflies. Photos courtesy of Matt Aubuchon, University of Florida.

upon emergence. Preparation of the holes in the dishes will take approximately 2 hours, and most dishes can be reused. Flies can be sexed at any point after emergence. Females can be separated into dishes and left for several hours prior to testing, if necessary. Substrates can be prepared and refrigerated for up to a week. The expected time commitment for placing flies in dishes is about a half hour the day of the experiments for a classroom of 25 students, after flies are separated and substrates prepared. Each lab should last approximately 45 minutes to 1 hour, depending on the experiment chosen and whether any modifications were made. Students who have finished all their replicates can assist the instructor to expedite cleanup time.

Experiment 1 Background & Preparation: Investigation on the Foraging Behavior of Gravid & Nongravid Houseflies

Gravid insects, or those with mature eggs, have theoretically satisfied their protein needs. The location of suitable oviposition sites may take precedence over foraging for a food source if protein needs are met, and gravid flies may spend less time on the experimental substrates, or may prefer substrates that are attractive for oviposition only. Conversely, flies that are no longer gravid will need to feed on protein to develop egg clutches. Both types of flies still rely on a carbohydrate source for energy, but the nutritional needs and behavior of each type of fly may vary in relation to the status of ovary development.

For this experiment, flies should be maintained for 4 to 5 days prior to the experiment with a diet of 67% granulated sugar, 27% powdered milk, and 6% powdered egg yolk (totaling ~100 g) in the cage, and this should be replaced three times a week. The day before the experiment, place a small amount of deli meat in a black cloth (about 10 × 10 cm) and secure with a rubber band. Soak this ball in hot water and place in a plastic cup in one of the cages. Flies should lay eggs, which look like elongated grains of rice, in this cup. Leave the cup for 4 to 6 hours before removing to ensure that all eggs have been laid. Eggs can be washed down the drain. This cage will be for the nongravid flies, and the remaining cage for the gravid flies.

Experiment 2 Background & Preparation: Investigation of the Effect of Previous Nutrition on Foraging Behavior of Houseflies

Upon emergence, adult females must locate a protein source to begin yolk deposition in developing eggs. Female houseflies have several cycles of ovarian development throughout their life span, and attraction to substrates may depend on fly age. Eggs are not produced when flies are fed only sucrose (Glaser, 1924); conversely, feeding on only protein and no carbohydrates results in a dramatically reduced life span and no eggs (Glaser, 1924; Turner & Hair, 1967). Foraging behavior may be related to nutritional needs based on the status of ovarian development as determined by the previous exposure to either protein or carbohydrates.

To test the influence of nutritional needs on young flies, after emergence, one cage of flies should be provided 100 g powdered egg yolk and the other cage 100 g granulated sucrose. Both cages of flies should have access to water. Flies should be held in these cages for 3 days before separating and starving females.

○ Student Experimentation

Materials & Methods for the Experiment

- Rolled gauze
- Substrates (suggested: blood, ham, honey, sugar water, Gatorade, etc.). Substrates should be liquid or pureed with water
- Paper towels and soap for cleanup
- Stopwatches
- Ice or access to a freezer
- Microscope (optional)
- Data-collection handout (Table 1)
- Water source
- Popsicle stick

Each student should have three dishes of flies from each variable, for a total of six (three gravid and three nongravid or three protein-fed and three sugar-fed, depending on the experiment chosen). Each group of students will conduct one full replicate per person, applying each treatment to each dependent variable and using a dry piece of gauze as a control (e.g., a group of six students will produce six complete replicates). Students should prepare their additional materials and be careful not to disturb or shake the flies. Each student should have six small rolls of gauze, a popsicle stick, the data table, and a stopwatch.

The instructor should demonstrate how to provide the substrate to the flies. The gauze strip should be rolled into a thin tube and dipped in the liquid substrate until saturated. Excess liquid can be squeezed out. The gauze strip should be inserted through the access hole with the popsicle stick or forceps. The gauze strip should remain perpendicular to the edge of the dish to ensure that the fly comes in contact with the strip.

Once the flies have been inserted into the dish, students should time each fly individually for 6 minutes. Each contact with the gauze is considered 1 second. If the fly contacts the gauze and remains on it longer than 1 second, students should use the stopwatch to count the total amount of time (in seconds) the fly remains on the gauze. Each group should calculate the average time each fly spent on each treatment and the standard deviation (SD).

○ Assessment

Following the experiment, the instructor led a class-wide discussion of the results. Some of the questions:

- Was your original hypothesis accepted or rejected?
- Did you have one treatment that was clearly better than the other?
- How can the physiological status of the housefly affect foraging behavior?
- What behaviors did you observe other than feeding?
- What do you think the flies were doing when they were not feeding?
- How is behavior related to disease transmission?
- How is behavior influenced by nutrition?
- Protein and sugar are necessary for nutrition, but what else would flies be attracted to?
- What could improve the experiment?
- What are some possible sources of error?
- Where would you go to look for literature related to this experiment to cite?

The instructor led a discussion of how these results, and what is already known about housefly nutrition and behavior, could be used to improve the control of houseflies. Baits are typically sugar based and include a lethal insecticide. Baits typically include an attractant (Leicht, 1993) and use the sugar granules to retain flies on the baits for a suitable time to allow ingestion but do not include a protein source. Students used critical thinking to synthesize how physiological status affects behaviors and to discuss what potential additives to bait may increase fly attraction, retention, and subsequent control.

Students in the introductory entomology laboratory were expected to write a lab report in the style of a refereed journal following this experiment. Students were graded on writing style, thorough methodology, correct analyis of data (Figure 4), critical thinking, and communication skills. Students were required to seek out and reference six additional peer-reviewed journal sources for their introduction and discussion sections, which introduced literature review as an important part of information synthesis.

At the conclusion, students were able to formulate a hypothesis, design and execute methods, and collect results. Students demonstrated

Table 1. Sample data-collecting table.

	Substrate Type (e.g., blood)	Time (seconds)					Mean	SD
		Rep. 1	Rep. 2	Rep. 3	Rep. 4	Rep. 5		
Protein								
Carbohydrate								
Control								

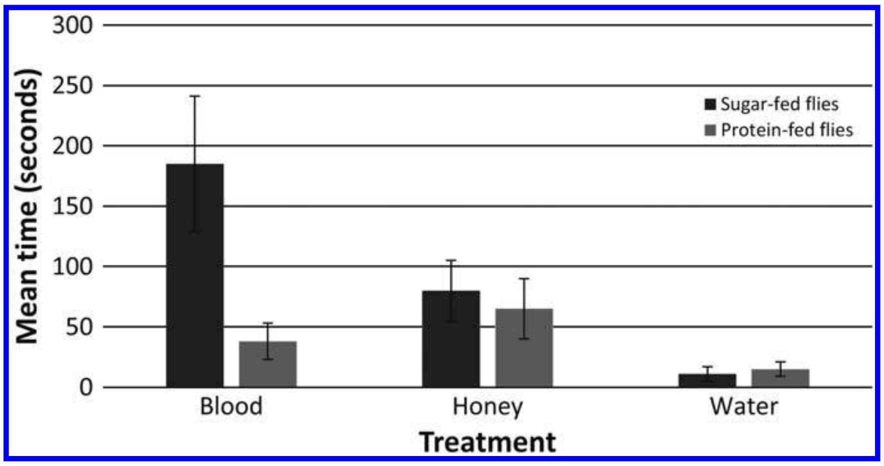

Figure 4. Example of student-generated histogram included as part of the results section of the laboratory report. Mean time (seconds ± SD) sugar-fed and protein-fed 4-day-old flies spent on each treatment (M. Rush, University of Florida).

information synthesis: 73.9% of students in the discussion section received a grade of 90% or better on this section. Students' ability to synthesize information was analyzed by assessing whether they compared the results from their experiments with relevant literature, whether the meaning of the results in a broader context was addressed, whether conclusions regarding the results and use of the data were made, and what improvements could be made to the methodology. A certain level of ingenuity was demonstrated by some students in formulating new ideas on fly control based on the collected data. However, a greater level of integration and interpretation of the results, relating the results to the big picture, was often difficult. It is recommended that enough time be spent on background to give students the necessary information. Additionally, following up the experiment with a class-wide or group-specific discussion on what the results could mean would facilitate a well-developed and thoughtful discussion section. Further analysis of student learning could be made with the addition of pretesting and posttesting to quantitatively assess learning gains.

Following the experiment and subsequent lab report, students were asked to provide feedback on their experience. About 75% of students who participated in the laboratory activity responded to the survey (n = 70). Approximately 75% of respondents answered that they enjoyed doing research in a laboratory setting with no known answer, as opposed to 25% who preferred to compare their results to known outcomes. Only 34% preferred to develop experiments including materials and methods based on background information, whereas the remaining 66% liked having a set way to proceed provided in class. This was an expected response, given the introductory level of the course and the previous scientific experience of the students as a whole. Approximately 72% of responding students preferred working in groups over working alone during the laboratory experiments, and 74% liked that the groups decided on the substrates to test, rather than the class as a whole, which gave each student greater influence on what was tested and the ability to have more comparisons made.

○ Conclusions

Inquiry-based and hands-on learning experiences like the experiments presented here can increase student performance, compared with confirmatory experiments, when teaching complex scientific concepts (Rissing & Cogan, 2009). Through this activity, students gained first-person experiences of scientific principles and learned to employ experimental methods to explore housefly feeding behavior as it is related to nutritional needs and how that might affect control. Students participated in guided-inquiry investigations with direct instruction from teachers to promote increased learning and develop scientific skills. Guided-inquiry experiments are better for evaluating evidence collected from student research, compared with confirmatory experiments or no experiments (Tien & Stacy, 1996).

Exposure to inquiry activities in the laboratory is a powerful learning tool and can shift students from being consumers of knowledge to being engaged in the production of knowledge (Neary, 2010). Students were committed to the experiment as a result of brainstorming methodology and choosing their own treatments, and they enjoyed working with live animals and unusual substrates such as blood and egg protein. Many students expressed gratitude that these experiments were not formulaic, with known results. Future classes would benefit from additional discussion following the discussion to put the results in context and lead to a more complete understanding of the process.

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