
ANIMAL BEHAVIOR

Background

The fruit fly *Drosophila melanogaster* is an organism commonly used in behavioral studies due to its short generation time and ease of culture. It is known to perform dozens of complex behaviors that can be quantified and explored, making it a perfect organism for studying animal behavior.

Animal behavior refers to the responses an animal makes and why it makes them. These behaviors are triggered by either internal or external stimuli and can be either an instinct (based on the organism's genes) or a learned behavior acquired by the interaction of the organism with its parents or surroundings.

Some of the simplest behaviors are those related to an organism's reaction to environmental factors such as light, sound, or moisture. If an organism changes its behavior in response to a stimulus but is not directed by the stimulus, it is called a *kinesis*. However, if the organism responds positively (moves towards) or negatively (moves away from) to the stimulus, the movement is called a *taxis*. For example, an organism might sense the presence of a chemical substance, and the organism may be attracted to the substance or repelled by it. The observed directional movements of the organism in response to the substance are referred to as *chemotaxis*.

In the lab, you will collect and analyze data from a choice chamber to identify whether fruit flies respond to an environmental stimulus and identify if taxis or kinesis behavior occurs.

Driving Question

How is the orientation behavior of fruit flies influenced by the presence of different stimuli in an environment?

Materials and Equipment

Use the following materials to complete the initial investigation. For experimenting with your design, check with your teacher to see what materials and equipment are available.

- Clear drinking straw
- Droppers (2)
- Cotton swabs (10)
- Timer
- Sheet of white paper
- Wingless fruit flies (10), or similar small organism
- Mashed ripe banana, 10 mL
- Mashed unripe banana, 10 mL
- Distilled water, 10 mL

Safety

Follow these important safety precautions in addition to your regular classroom procedures:

- Wear safety goggles when working with chemicals.
- Students should not eat any food items in the lab.
- Treat all living organisms with care.

Initial Investigation

Complete the following investigation before designing and conducting your experiment. Record all observations, data, explanations, and answers in your lab notebook.

1. Place 5–10 drops of distilled water onto the end of a cotton swab and place the swab into one end of a clear drinking straw.
2. Using a paper funnel, place 10 wingless fruit flies into the drinking straw.
3. Place 5–10 drops of distilled water onto a second dry cotton swab and place it into the other end of the drinking straw.
4. Lay the choice chamber with the fruit flies onto a white sheet of paper. Label one end of the straw as side A and one as side B. Allow the fruit flies 5 minutes, with no disturbances, to acclimate to the choice chamber.
- ❓ 5. What is the purpose of collecting data for a situation in which fruit flies are given the same choice on either side of the choice chamber?
6. After the 5 minute acclimation period, start a 5 minute observation period. Count the number of flies on side A and the number of flies on side B every minute for 5 minutes. Record your observations in a data table arranged in Table 2.
- ❓ 7. For the following situations using the choice chamber, predict whether you expect fruit flies to exhibit a preference or whether you predict the null hypothesis to be supported (that the flies will have no preference).
 - (a) ripe bananas vs. distilled water
 - (b) ripe bananas vs. unripe bananas
8. Using the same choice chamber and fruit flies, repeat the above procedures to expose the flies to the two combinations of substances specified in the previous step. Record the data in your lab notebook for each one.
9. Calculate the average number of flies on each side of the chamber for each situation.

10. Complete a chi-square analysis of the results to determine if the flies' distribution in the choice chambers is significant. (Table 1 is provided for reference.)

Use this null hypothesis for all experiments: The fruit flies do not have a preference for either substance in the choice chamber.

Table 1: Chi-square distribution

Degrees of Freedom	Probability p Value					
	0.75	0.50	0.25	0.10	0.05	0.01
1	0.10	0.46	1.32	2.71	3.84	6.64
2	0.58	1.30	2.77	4.60	5.99	9.21
3	1.21	2.37	4.11	6.25	7.82	11.34
4	1.92	3.36	5.39	7.78	9.49	13.28

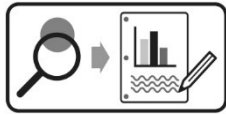
Table 2: Determination of significant trends

Time (minute)	Number of Fruit Flies					
	Water vs Water		Water vs Ripe Banana		Ripe vs Unripe Banana	
	A	B	A	B	A	B
1						
2						
3	RECORD ANSWERS & DATA IN YOUR					
4	NOTEBOOK.					
5						
Total (Observed o)						
Average(Observed o)						
Expected e						
$(o - e)^2/e$						
$\chi^2 = \sum[(o - e)^2/e]$						

11. After reviewing the data and completing a chi-square test, what is your conclusion? Did the fruit flies demonstrate a chemotaxis to any of the substances that were tested? Was it a positive or negative taxis?

Design and Conduct an Experiment

Organisms exhibit a variety of behaviors that can be classified as taxis or kinesis behavior. Using a choice chamber and a small organism, select an environmental factor and conduct an experiment to determine if that factor produces a taxis or kinesis behavior in the organism.



Design and carry out your experiment using either the Design and Conduct an Experiment Worksheet or the Experiment Design Plan. Then complete the Data Analysis and Synthesis Questions.

Design and Conduct an Experiment: Data Analysis

1. From your observations and your data:
 - a. Describe how the independent variable you manipulated affected the behavior of the wingless fruit flies. Does chi-square analysis of your data indicate the null hypothesis can be rejected? Justify your claim with evidence from your experiment.
 - b. Based on the evidence you collected, explain why the results occurred.
2. Is there any evidence in your data or from your observations that experimental error or other uncontrolled variables affected your results? If yes, is the data reliable enough to determine if your hypothesis was supported?
3. Identify any new questions that have arisen as a result of your research.

Synthesis Questions

1. Animal adaptive behaviors are crucial for survival. Two types of adaptive behaviors are *innate behaviors* and *learned behaviors*. Explain how these two behaviors can impact an individual organism.
2. Animals exhibit behaviors that are classified as either a *taxis* or *kinesis*. What is the difference between taxis and kinesis in relation to animal behavior? In addition to chemotaxis, what other types of taxis responses exist?
3. An experiment was performed to investigate aggressive behavior in olive fruit flies (*B. oleae*).¹ Aggressive behavior was observed in swarms of flies around olive trees—males fighting to occupy leaves to perform courtship displays and females fighting for sites for laying eggs. Additionally, both sexes could gain access to food sources by occupying leaves or fruits on the tree. Investigators recorded fly behavior with high-speed video cameras and determined three behaviors to categorize as aggressive: wing waving, fast running toward the opponent, and pouncing and boxing on the head and thorax of the foe. One of the driving questions of the investigation was: Do resident flies win more combats than non-resident flies?

NOTE: A resident fly is a fly placed into the chamber first, allowing it to establish a territory (“residence”) before other flies are added to the testing chamber. A “win” is awarded to a fly if it remains on an olive leaf for at least 30 seconds after an aggressive interaction that displaces another fly.

¹ Benelli, G. Aggressive Behavior and Territoriality in the Olive Fruit Fly, *Bactrocera oleae* (Rossi) (Diptera: Tephritidae): Role of Residence and Time of Day. *Journal of Insect Behavior* (2014) 27:145–161. doi 10.1007/s10905-013-9411-7.

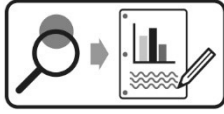
Table 3: Observations of aggressive interactions in olive fruit flies

Sex	Initiator of an Aggressive Interaction		Winner of an Aggressive Interaction	
	Resident	Non-resident	Resident	Non-resident
Males	16	14	21	9
Females	19	11	22	8

- a. Write a null hypothesis for the driving question of the experiment and use chi-square analysis to determine if the null hypothesis should be accepted or rejected. Are the results different for different sexes?
- b. Aggressive behavior has a genetic basis and has been conserved in insect evolution. In other words, the behavior is common in many insect taxa. Explain the relationship between natural selection and behavior in organisms.

Design and Conduct an Experiment Worksheet

Organisms exhibit a variety of behaviors that can be classified as taxis or kinesis behavior. Using a choice chamber and a small organism, select an environmental factor and conduct an experiment to determine if that factor produces a taxis or kinesis behavior in the organism.



Develop and conduct your experiment using the following guide.

1. Based on your knowledge of animal behavior, what environmental factors (abiotic or biotic) could affect an organism's behavior?

2. Create a driving question: choose one of the factors you've identified that can be controlled in the lab and develop a testable question for your experiment.

3. What is the justification for your question? That is, why is it biologically significant, relevant, or interesting?

4. What will be the independent variable of the experiment? Describe how this variable will be manipulated in your experiment.

5. What is the dependent variable of the experiment? Describe how the data will be collected and processed in the experiment.

6. Write a testable hypothesis (If...then...). Is this hypothesis the null hypothesis or alternate hypothesis for the chi-square analysis?

7. What conditions will need to be held constant in the experiment? Quantify these values where possible.

8. How many trials will be run for each experimental group? Justify your choice.

9. What will you compare or calculate? What analysis will you perform to evaluate your results and hypothesis?

10. Describe at least 3 potential sources of error that could affect the accuracy or reliability of data.

11. Use the space below to create an outline of the experiment. In your lab notebook, write the steps for the procedure of the lab. (Another student or group should be able to repeat the procedure and obtain similar results.)

12. Have your teacher approve your answers to these questions and your plan before beginning the experiment.

