ADVANCED WORKSHEET STUDENT HANDOUT

# THE VIRTUAL STICKLEBACK EVOLUTION LAB

### INTRODUCTION

As you complete each part of the virtual lab, as assigned by your teacher, answer the questions below in the space provided.

### QUESTIONS

### INTRODUCTION

- 1. Define model organism.
- 2. How do the spines protect ocean stickleback fish?
- 3. Why do researchers think that freshwater stickleback populations arose when ancestral ocean-dwelling stickleback became trapped in freshwater lakes?
- 4. *Watch the video about pelvic reduction in freshwater stickleback.* The loss of stickleback pelvic spines is similar to the loss of which body parts in some four-legged vertebrates?
- 5. From a researcher's perspective, what is the advantage of having access to hundreds of postglacial lakes?
- 6. Watch the video with evolutionary biologist Dr. Michael Bell.
  - a. According to Dr. Bell, what insights can we gain from studying modern populations of organisms?
  - b. What insights can we gain from studying the fossil remains of populations of organisms?
- 7. *Watch the video with Dr. David Kingsley explaining hind-limb reduction.* Name two other vertebrate animals whose evolutionary histories included the loss of hind limbs.
- 8. Provide two reasons why the threespine stickleback fish is a useful model organism for studies in evolution.

### OVERVIEW

1. What is the difference between marine, sea-run, and freshwater stickleback fish? Be specific.

### 2. Watch the stickleback fish anatomy video.

- a. What are the two types of paired fins?
- b. How are stickleback pelvic fins different from those of other teleost fish species? (Teleosts are a large group of ray-finned fish, including, for example, minnows, tuna, and goldfish.)

### c. Dichotomous (circle one choice per pair that best completes the following statements):

- 1. Anterior means toward the (head / tail) of the fish while posterior toward the (head / tail) of the fish.
- 2. (Dorsal / Ventral) means the back side of the fish and (dorsal / ventral) is the belly side.
- d. What advantage does the ability to simultaneously lock in place the two pelvic spines with the dorsal spine directly above them provide to the stickleback?

### **TUTORIAL 1**

- 1. Describe the following scoring categories used in the virtual lab:
  - a. Complete pelvis:
  - b. Reduced pelvis:
  - c. Absent pelvis:
- 2. Scroll down the Overview page and watch the short video on pelvic scoring. Does the pelvis of every fish with pelvic reduction look the same? What are the similarities and differences?

## 3. Complete Tutorial 1.

### **EXPERIMENT 1**

- 1. What is the overall objective of Experiment 1?
- 2. Click on the link to the map of Alaska, and then click on the blue pin "A" on the larger map. Which lake is located between Bear Paw Lake and Frog Lake just to the north?
- 3. In a population, what happens to organisms that are better adapted to the environment in which they live?
- 4. *Watch the short video on how postglacial lakes form.* What do researchers think is one of the selective pressures for the complete pelvis trait in ocean-dwelling threespine stickleback fish?

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# The Making of the Fittest: Evolving Switches, Evolving Bodies

- 5. *Watch the short video on the Cook Islet scenery.* What features in the various lake habitats might be selecting for either a reduced or complete pelvis?
- 6. In the virtual lab window, complete Part 1: Staining the Fish.
- 7. Why do you think it is important to empty the used stain, destaining solution, and water under a fume hood?
- 8. In the virtual lab window, proceed to Part 2: Scoring the Fish.
- 9. In the window on the right, click on the Random Sampling link. What is a **population**?
- 10. Why are random samples used to study populations?
- 11. What is one advantage of studying larger-sized samples?
- 12. Give an example of **sampling bias**.

### 13. Watch the video of Dr. Bell explaining sampling in Bear Paw Lake.

- a. Why is variation in a population critical to evolution?
- b. From his 20 years of studying Bear Paw Lake, is there evidence that stickleback pelvic structures have changed in this population within this time period?
- c. Is Bear Paw Lake similar to other lakes Dr. Bell has studied?

### 14. Complete Part 1 of the lab in the window on the left.

- 15. Why is it important that the labels included in the specimen jars be made of special paper that does not disintegrate in alcohol over time?
- 16. Examine the pelvic score data you just collected. Does pelvic phenotype differ between Bear Paw Lake and Frog Lake fish? Explain.

### 17. Complete the graphing exercise as instructed by your teacher.

18. After graphing, verify your data. How do your data compare to those obtained by Dr. Bell's lab?

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- 19. **Complete Experiment 1 Quiz. Take time to read the explanation for each correct choice provided** *following each question.*
- 20. Explain why the stickleback fish in Frog Lake are more similar to ocean and sea-run stickleback than they are to the stickleback fish in Bear Paw Lake.
- 21. In addition to predators, can you think of other environmental factors that might be responsible for the differences between Bear Paw Lake and Frog Lake stickleback populations? How would you test your prediction?

### 22. Upon completion of the quiz, click on Experiment 1 Analysis.

- 23. In this chi-square analysis, what is your null hypothesis?
- 24. Complete the following tables as you perform the chi-square calculations in the lab.

Bear	Paw	Lake

Phenotype	Observed (o)	Expected (e)	(о-е)	(o-e) <sup>2</sup>	(o-e)²/e
	Total =	Total =			χ <sup>2</sup> =

### Frog Lake

Phenotype	Observed (o)	Expected (e)	(о-е)	( <b>o-e</b> ) <sup>2</sup>	(o-e)²/e
	Total =	Total =			χ <sup>2</sup> =

Morvoro Lake

Phenotype	Observed (o)	Expected (e)	(o-e)	(o-e) <sup>2</sup>	(o-e)²/e
	Total =	Total =			χ <sup>2</sup> =

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- 25. For each lake's chi-square calculation, how many degrees of freedom are there? \_\_\_\_\_\_ How did you arrive at this number?
- 26. What are the p values for each lake?
  - a. Bear Paw Lake
  - b. Frog Lake
  - c. Morvoro Lake \_\_\_\_
- 27. Do you reject or fail to reject your null hypothesis for each lake?
  - a. Bear Paw Lake \_\_\_\_\_
  - b. Frog Lake
  - c. Morvoro Lake \_\_\_\_\_
- 28. Complete the Chi-Square Analysis Quiz. Take time to read the explanation for each correct choice provided following each question.
- 29. Explain what it means to reject the null hypothesis.
- 30. Explain what it means to fail to reject the null hypothesis.
- 31. Explain the difference between the results of the chi-square calculations for Bear Paw Lake and Morvoro Lake.

## 32. Proceed to Tutorial 2.

# TUTORIAL 2

- 1. What score would you assign to a fossil specimen that has only one pelvic spine visible?
- 2. Some stickleback fossils show no signs of pelvic structures. What are some **possible sources of error** associated with scoring a stickleback fossil "absent"?

## 3. Watch the video about scoring stickleback fish fossils.

4. Complete Tutorial 2.

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# The Making of the Fittest: Evolving Switches, Evolving Bodies

### **EXPERIMENT 2**

- 1. What is the overall objective of Experiment 2? Explain what the information collected in Experiment 2 will allow you to estimate.
- 2. *Watch the short video on the Truckee Formation in Nevada.* What does each sedimentary rock layer of the Truckee Formation represent?
- 3. What is one type of information that researchers can gain from fossils that they cannot get from living populations?
- 4. Begin the Experiment in the window on the left. Watch the video showing Dr. Bell preparing a fossil.
- 5. In the virtual lab window, complete Part 1: Preparing Fossils.
- 6. Proceed to Part 2: Scoring Fossils. In this virtual lab, you will graph fossil data from six rock layers (your data came from two of these layers). Approximately how many years apart are any two adjacent samples?
- 7. You collected data on pelvic structures using fossils from rock layers 2 and 5. Approximately how many years of deposition separate these two layers?
- 8. Which layer is older, 2 or 5? Explain your answer.

### 9. Complete Part 2 of the lab in the window on the left.

10. When you obtained your pelvic phenotype totals, do the fossils in layer 2 differ from those in layer 5? Explain how.

### 11. Complete the graphing Experiment as instructed by your teacher.

- 12. After graphing, verify your data. How do your data compare to those collected by Dr. Bell and colleagues?
- 13. **Complete Experiment 2 Quiz. Take time to read the explanations for each correct choice provided** *following each question.*
- 14. What can be inferred about the presence or absence of predatory fish when the Truckee Formation was a lake? Explain the evidence for your inference.

### 15. After completing the quiz, click on Experiment 2 Analysis.

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16. Complete the tables below as you perform the rate calculations. (The link to the instructions is very helpful.)

Sample Layer	Number of Fish with a Complete Pelvis	Total Number of Fish Sampled	Relative Frequency of Complete Pelvis Trait in Population Sampled
1			
2			
3			
4			
5			
6			

Time	Decrease in Percentage of Complete Pelvis Trait per Thousand Years (Rate of Change)
First 3,000 years	
(Layer 1 to Layer 2)	
Next 3,000 years	
(Layer 2 to Layer 3)	
Next 3,000 years	
(Layer 3 to Layer 4)	
Next 3,000 years	
(Layer 4 to Layer 5)	
Next 3,000 years	
(Layer 5 to Layer 6)	
Total 15,000 years	
(Layer 1 to Layer 6)	

- 17. What does it mean when the rate of change is a negative number?
- 18. Complete the Analysis Quiz. Take time to read the explanation for each correct choice provided following each question.
- 19. Describe the trend in the data over time.
- 20. Why is it important to calculate the rate of change over time?
- 21. In what ways is the change in the complete pelvis phenotype in the fossils from the Nevada lakebed similar to what might have occurred in Bear Paw Lake from Experiment 1?

### 22. Proceed to Experiment 3.

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# **EXPERIMENT 3**

- 1. What is the overall objective of Experiment 3?
- 2. What is one function of the *Pitx1* gene?
- Watch the video segment from the HHMI short film, "Evolving Switches, Evolving Bodies."
  a. What is the relationship between changes in body form, the process of development, and genes?
  - b. Why did Dr. Kingsley do genetic crosses with stickleback fish?
- 4. In the virtual lab window, click on the blue gloves and perform Part 1: Staining the Fish (you can skip the staining if you completed it in Experiment 1). Proceed to Part 2: Scoring Pelvic Asymmetry.
- 5. Which one of the three pelvic phenotypes is analyzed in more detail in this Experiment?
- 6. What is the difference between *left-biased* and *right-biased* pelvic asymmetry?
- 7. In the virtual lab window, complete Part 2: Scoring Pelvic Asymmetry.
- 8. Complete the graphing exercise as instructed by your teacher.
- 9. After graphing, verify your data. How do your data compare to those collected by Dr. Bell and colleagues?
- 10. **Complete Experiment 3 Quiz. Take time to read the explanation for each correct choice provided** *following each question.*
- 11. The pelvic asymmetry observed in stickleback fish from Bear Paw Lake and in Coyote Lake is biased toward which side?
- 12. Based on previous research conducted mice, what do the findings on pelvic asymmetry from Bear Paw Lake and Coyote Lake stickleback suggest about the genetic mechanisms underlying pelvic reduction?

13. When you are finished with question 5 of the quiz, watch the video of Dr. Bell explaining his findings. (If you have already clicked to the summary page, you can simply click the "back" arrow at the top of the computer window and see the link to this video.)

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# 14. After completing the quiz, click on Experiment 3 Analysis.

15. For this chi-square analysis, what is the null hypothesis?

# 16. Complete the following tables as you perform the chi-square calculations.

Bear Paw Lake	
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Phenotype	Observed (o)	Expected (e)	(о-е)	(o-e) <sup>2</sup>	(o-e)²/e
	Total =	Total =			χ <sup>2</sup> =

## Coyote Lake

Phenotype	Observed (o)	Expected (e)	(o-e)	( <b>o-e</b> ) <sup>2</sup>	(o-e)²/e
	Total =	Total =			χ <sup>2</sup> =

17. For each lake's chi-square calculation, how many degrees of freedom are there?

- 18. What are the p values for each lake?
  - a. Bear Paw Lake
    - b. Coyote Lake \_\_\_\_\_
- 19. Do you reject or fail to reject your null hypothesis for each lake?
  - a. Bear Paw Lake \_\_\_\_\_
  - b. Coyote Lake \_\_\_\_\_
- 20. Complete the Chi-Square Analysis Quiz. Take time to read the explanation for each correct choice following each question.
- 21. What does it mean to reject the null hypothesis?
- 22. Name two factors that could be contributing to left bias in a stickleback with pelvic reduction.

## 23. Complete the Final Quiz.

24. Print the final Summary page if instructed to do so by your teacher.

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- 25. Listed below are the key concepts of this virtual lab. Write a short essay explaining how this virtual lab provides **evidence** for **EACH** of the key concepts.
  - Natural selection can drive the evolution, not just of simple traits like coat color or body size, but also of complex traits like the size and shape of skeletons.
  - Different environments with different predators, food sources, or resource limitations apply different selective pressures on the shape of animal bodies.
  - Important evolutionary insights can be gained by making comparisons among fossils of different ages and locations and among living populations under different selective pressures, and by comparing fossils to living populations.
  - Careful quantitative analysis of the traits of living populations can tell us about mechanisms of selection, including genetic mechanisms, whereas quantitative analysis of populations of fossilized specimens can provide a record of change over time and even the pace of specific adaptive changes.
  - Statistical analysis is essential for gauging confidence in conclusions drawn from population data, which can include natural variation, populations in transition, and measurement error by the researcher.

#### AUTHOR

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