INQUIRY & INVESTIGATION

Modeling Evolution in the Classroom: The Case of Fukushima's Mutant Butterflies

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Abstract

New science standards and reform recommendations spanning grades K–16 focus on a limited set of key scientific concepts from each discipline that all students should know. They also emphasize the integration of these concepts with science practices so that students learn not only the "what" of science but also the "how" and "why." In line with this approach, we present an exercise that models the integration of fundamental evolutionary concepts with science practices. Students use Avida-ED digital evolution software to test claims from a study on mutated butterflies in the vicinity of the compromised Fukushima Daiichi Nuclear Power Plant complex subsequent to the Great East Japan Earthquake of 2011. This exercise is appropriate for use in both high school and undergraduate biology classrooms.

Key Words: Evolution; digital evolution; mutation; mutation rate; model.

Science education in the United States is changing rapidly. New national reforms for grades K–16 all have a common goal: to focus on a handful of key concepts and integrate these with science practices (AAAS, 2011; College Board, 2011; NGSS Lead States, 2013). Implemented as intended, these reforms have the potential to fundamentally change the way that science is taught – ultimately,

it is hoped, producing a new generation of learners who engage in critical reasoning and who understand the nature and processes of science more deeply than ever before. At the same time, the Association of American Universities (2011) has launched a 5-year initiative to improve the way that science is taught at the undergraduate level, particularly in introductory courses, where the most attrition occurs. In order to accomplish these reform-

oriented changes, science instructors will need new tools and materials that will help them create courses that are aligned with reform recommendations.

Integrating disciplinary core ideas, crosscutting concepts, and science practices (National Research Council, 2011; NGSS Lead

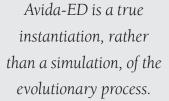
States, 2013) will be inherently more challenging for certain subject areas than for others. For example, phenomena that occur in deep time, such as the shifting of tectonic plates or the evolution of biological organisms, have proved difficult to represent in the classroom (Trend, 2001; Catley & Novick, 2009). A solution to this problem for evolutionary biology is Avida-ED, evolution software that models biological evolution and allows students to engage in authentic science practices while learning evolution and genetics content. Unlike other currently available evolution education software, Avida-ED is a true instantiation, rather than a simulation, of the evolutionary process (Pennock, 2007b). The digital organisms, called Avidians, replicate their circular genomes (strings of computational instructions) and pass them to their offspring. Mistakes, in the form of random substitutions, are made during replication, producing genetic variation among individuals. Accumulation of these mutations can result in the ability of the organisms to perform simple computational functions, and these are rewarded by the virtual environment in the form of "energy" - that is, the ability to replicate faster. Organisms that replicate more quickly will tend to leave behind more offspring. As a result, populations of Avidians evolve over time through Darwinian natural selection. When using Avida-ED, students witness evolution

> in action while developing hypotheses and designing experiments to test various aspects of the evolutionary mechanism in real time (Pennock, 2007a; National Research Council, 2012).

• Why Digital Evolution?

Avida-ED is an educational version of the Avida digital evolution research platform cur-

rently being used by evolutionary biologists to conduct "impossible" evolutionary experiments – those that cannot, because of practical limitations, be accomplished with biological organisms. Avida has been used in dozens of publications to test a wide range of biological hypotheses, ranging from how evolution produces observed patterns



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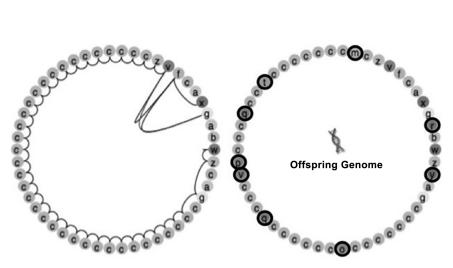


Figure 1. The circular Avidian genome contains 50 instructions (dots with letters, 26 different instructions total). The ancestor (left) has only the ability to copy itself. The offspring genome (right) was produced by replicating the ancestor at a 15% mutation rate. Mutated instructions are circled in bold.

from complex features (Lenski et al., 2003) to how cooperative behaviors evolve (Clune et al., 2011). Avida makes this possible by producing an instance of the evolutionary process in a digital world, in a fraction of the time it takes for the same process to occur in nature.

Avida-ED has the power of a research platform but features a user interface that allows easy manipulation of the environment (e.g., dish size and available resources) and of the organisms (e.g., mutation rate; see Figure 1). By adjusting environmental settings, students can use Avida-ED to design experiments that test hypotheses they have developed. They collect, analyze, and graphically represent data and communicate their findings with others. Avida-ED has other advantages that make it especially useful for discovery-based learning in small or large classes. Multiple experimental trials can be conducted in a single class period, resulting in the collection of large amounts of data. Unlike biological organisms, Avidians require no material resources, and the program can be downloaded without charge (see Notes below). Students witness evolution in action, learn about fundamental evolutionary concepts, and engage in authentic science practices, all within the limited space and time of the typical classroom (Speth et al., 2009).

○ Guided Inquiry Activity

Context: Aftermath of the Great East Japan Earthquake

On 11 March 2011, the largest earthquake ever to hit Japan, and one of the five largest in recorded history, occurred 70 km off the coast of Tōhoku. The quake triggered a tsunami that produced waves with heights ≤ 40.5 m that traveled ≤ 10 km inland, causing extensive damage to property and significant loss of human life. Among the infrastructure casualties was the Fukushima Daiichi Nuclear Power Plant complex. Three reactors sustained heavy damage, resulting in the worst nuclear accident since Chernobyl in 1986. The area within a 30-km radius of the Fukushima plant was determined to have dangerously high levels of leaked radiation, with the highest levels ≤ 3 km from the plant. The Japanese government prohibited access to this area and ordered the evacuation of anyone living ≤ 20 km from the plant. People living 20–30 km away were put on high alert and also encouraged to evacuate (Black, 2011).

The disaster at Fukushima has provided scientists with a unique opportunity to investigate the biological impact of radiation. In particular, a butterfly species, the pale grass blue (Zizeeria maha), is helping researchers pursue questions about the immediate and long-term effects of acute and chronic exposure to radiation at various doses (Hiyama et al., 2012). Butterflies collected closer to the power plant experienced larger doses of radiation than those farther away. Exposure to larger doses was associated with increased infertility, mortality, and incidence of physiological abnormalities. Many of these abnormalities were inherited and amplified in offspring of butterflies that had been exposed to the radiation initially as overwintering larvae.

The researchers concluded that "it is most likely that the abnormal phenotypes observed are produced by random mutations caused by the exposure to radiation" (Hiyama et al., 2012, p. 8). That is, butterflies that were located closer to the source of radiation (the damaged reactors) received larger doses and experienced increased mutation rates; as a result, they experienced more random mutations, including harmful ones, that were responsible for the higher incidence of abnormal phenotypes. This pattern – an increasing mutation rate produces a greater number of random mutations, resulting in abnormal phenotypes – is what students will test in Avida-ED.

Fundamental Evolutionary Concepts: Mutations & Mutation Rates

The basic elements of the Darwinian algorithm for natural selection include variation, inheritance, and selection. In order for students to understand the process of evolution (and move on to more advanced aspects of the underlying mechanism), they must first master these fundamental concepts. It is therefore important that students understand both mutation and mutation rate in order to make sense of population variation that will be subjected to environmental selection, resulting in evolutionary change. In the current exercise, students observe that mutations occur randomly as mistakes during genome replication, that they can be either deleterious or neutral (they will likely not see beneficial mutations during this exercise), and that the number of mutations, including harmful mutations, will increase as the mutation rate increases, resulting in a larger proportion of adverse genetic effects.

Activity Walk-through

Materials required for this lesson include computers with Avida-ED installed, the source article (Hiyama et al., 2012), which is freely available for download from the publisher's website, and a handout containing procedural information, questions, and space for recording data. Students are expected to have read the article and the handout before coming to class. At the start of the class period, the instructor facilitates a brief discussion of the article, summarizing the main patterns and claims. Next, the instructor tells the students that they will use digital organisms to model the conditions in the paper,

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in order to test the claim that the phenotypic abnormalities observed in butterflies were caused by exposure to radiation from the damaged nuclear power plant. The instructor will briefly introduce Avida-ED, demonstrating in the Organism viewer how mutations occur during genome replication and explaining how natural selection of accumulated mutations by an environment can allow organisms to perform simple functions. Then, following the handout, students develop a null hypothesis and one or more alternative hypotheses about the effects of mutation rate on genome replication. They should make predictions about what will happen to an individual organism's genome as it replicates at different mutation rates. Students should take a few minutes to answer these questions on their own, and then consult with a neighbor before sharing with the entire class (think-pair-share). The instructor should keep track of student predictions in a public space (board, overhead, etc.) where they can be referred to later.

In small groups, one group per computer, students replicate individual Avidians under different experimental conditions. In the Organism viewer, students click and drag a provided organism from the freezer into the viewing pane and observe it replicate, noting the functions it performs. Then, under the Settings tab, they set the per site mutation rate to 1% and follow the procedure as described in the handout: replicate the ancestor 10 times, setting up a new run each time; using the data sheet provided, record the number of mutations in each offspring; note how each offspring differs from the ancestor by recording which specific instructions in the genome have changed; and save each offspring to the freezer. After saving all 10 offspring organisms, students will drag each saved offspring, one at a time, from the freezer into the Organism viewing pane to examine its genome and determine which of the 10 functions (9 "metabolic" functions plus the ability to replicate), if any, each organism can execute. They should record the number of lost functions alongside the number of mutations for each replication event. This entire process should then be repeated at mutation rates of 5%, 10%, and 15%. When the students are finished, they may combine their data to create a single class data set; doing so makes patterns clearer to see.

As students report their results, the instructor should graph the combined class data, showing the average number of mutations and abnormality rate – calculated as the percentage of functions lost from the total – for each of the four mutation rates (see sample data in Table 1 and Figure 2 as examples). Students should then decide which of their earlier predictions best matches the patterns shown by the data.

Finally, the instructor engages the students in discussion about how well their data support the researchers' claim that the increase in abnormality rate is accounted for by an increase in random mutations

Table 1. Sample data. The number of mutations (substitutions in offspring genome subsequent to replication of the ancestor genome) and number of abnormalities (functions lost in offspring) were recorded for offspring organisms produced from replicating the same ancestor organism 10 times at each of 4 mutation rates.

	Mutation Rate							
	1%		5%		10%		15%	
Offspring	Mutations (n)	Abnormalities (n)	Mutations (n)	Abnormalities (n)	Mutations (n)	Abnormalities (n)	Mutations (n)	Abnormalities (n)
1	0	0	2	10	3	10	11	10
2	1	1	1	10	1	1	10	7
3	0	0	2	3	4	3	6	3
4	1	0	2	10	5	10	8	10
5	1	10	2	10	2	6	8	10
6	2	6	1	2	2	9	8	10
7	0	0	0	0	5	10	9	10
8	3	0	1	9	4	10	5	10
9	1	0	4	10	6	10	8	10
10	0	0	2	10	4	0	5	10
	Average Number of Mutations	Abnormality Rate						
	0.9	17%	1.7	74%	3.6	69%	7.8	90%



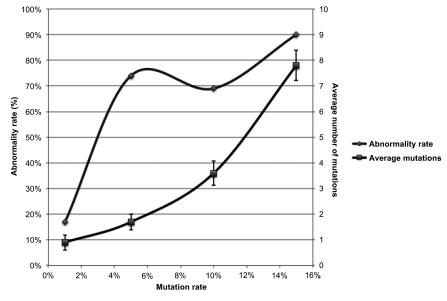


Figure 2. Graph of sample data from Table 1, depicting relationships between mutation rate and abnormality rate (r = 0.85) and between mutation rate and average number of mutations (r = 0.96). Error bars represent the standard error of the mean.

due to level of radiation exposure. This discussion can be for the purposes of formative assessment.

Summary of Patterns: The Effect of Mutation Rates on Individuals

As mutation rates increase, so do the number of mutations per replication event and the number of physiological abnormalities (Figure 2). This relationship can be explained by an increase in the number of harmful mutations that occur at higher mutation rates. In Avida-ED, this means that as mutation rates increase, there is a greater probability that the genes (i.e., sequences of instructions) coding for various phenotypes (here, ability to perform the 9 metabolic functions and to replicate) will become broken during genome replication. In Z. maha, it means that there is an increased incidence of infertility, mortality, and abnormal phenotypes, just as Hiyama et al. (2012) reported. Students can compare a graph of their data to the figures provided in that article (especially fig. 4b, which shows a positive correlation between abnormality rate and ground radiation dose, which serves as a proxy for mutation rate; p. 5). Thus, the model system independently demonstrates the pattern found in the study, supporting the researchers' claim.

O Extensions

This lesson was written to be used in an undergraduate biology course but could easily be adapted for use in a high school or AP biology class. The exercise is designed to engage students in a wide range of authentic scientific practices, from reading primary scientific literature to hypothesis testing to evidence-based argumentation, and these can be limited or augmented, depending on how the instructor chooses to use the materials. In addition, we have provided in the lesson materials suggestions for supplemental activities to provide additional scaffolding, as well as variations for making the lesson more or less guided or learner-centered. Examples of variations include the following:

- Instead of asking students to send their data, the instructor may choose to use the sample data provided.
- The instructor can give the collected class data set (or sample data) to students/groups and ask them to decide how to analyze it, or provide them with directions on how to analyze it.
- The instructor can establish the problem as described (testing the scientists' claim) and ask students to come up with an experimental protocol.
- The instructor can give the students just the initial problem (description of the disaster) and ask them to predict the effects on organisms in the vicinity of the nuclear power plant. They can then test their predictions in Avida-ED and later compare the results of their investigations to the findings in Hiyama et al.'s (2012) report.

O Addressing Misconceptions

We recommend that students be introduced to this activity only after they have been introduced to the idea of genetic variation due to random mutation. Instructors should be prepared to address potential misconceptions that are commonly associated with mutations, such as the idea that mutations are always harmful. This may be especially important to address given that the Fukushima butterfly exercise focuses specifically on the deleterious effects of mutations. The instructor may wish to discuss the random nature of mutations and point out that they occasionally have beneficial effects or have no effect at all (in the case of "silent" or neutral mutations), and even encourage students to find examples of these in their own data (Table 1). Additionally, in order to understand the concept of mutation rate, it is helpful if students have a basic knowledge of probability. If they do not, they may struggle with what is meant by a particular mutation rate. For example, rather than indicating that there will always be a 10% difference between the ancestor and offspring, a 10% mutation rate means that each site in the genome independently has a 10% chance of changing during replication. The actual number of mutations in a given replication event will vary according to a normal distribution (this can also be confirmed through examination of data collected during the exercise; Table 1). Other exercises on the Avida-ED website deal specifically with these concepts and may be used in conjunction with the current activity at the instructor's discretion.

○ Conclusion

This exercise is one example of how digital evolution allows instructors to integrate biological course content about evolution with authentic engagement in scientific practices. It gives students the opportunity to test claims made by established scientists in a peer-reviewed

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research article. It allows students to observe evolutionary processes in action and to engage in a variety of authentic scientific practices to see how scientific hypotheses can be tested by models. Using digital evolution in this way, instructors are able to combine course content with practices. We are developing a suite of such exercises to help students overcome a range of common misconceptions about evolutionary processes and to have them experience for themselves how science is done. Our hope is that these flexible exercises demonstrate a viable and scalable model for how instructors might restructure their own pedagogy to better align with reform recommendations.

O Notes

All materials, as well as additional information and resources, are available free of charge for download from http://avida-ed.msu.edu. A pre-evolved organism for this lesson is also available for download from the Avida-ED website, but instructors may evolve one themselves or have students use one from some previous exercise – the only requirement is that the ancestor organism for this exercise is viable (able to replicate) and able to perform all nine metabolic functions.

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