Glowing Friends

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SRE 2018
Curriculum plan

Contents

Author's Note

I have several ideas for the development of my curriculum based on Vibrio fischeri. This will be based on articles about scuba diving at night in Hawaii, glowing fish salad and infection.

1. Hook for the students: pipetting V. fischeri cells in growth broth into well plates; shaking overnight and observing the changes (bioluminescence and microscopy)
2. Homework: Read up on V. fischeri and its symbionts (fish and Hawaiian squid)
3. Determine growth rate of two strains of V. fischeri (fish and squid symbionts) using spectrophotometer.
4. Effect of antoinducers on luminescence.
5. Bioinformatics: using BLAST to determine genetic homology between the 2 strains and different species of Vibrio (infectious and noninfectious). Creating cladogram usng these organisms.

Stories
https://www.nationalgeographic.com/travel/florida-land-and-sea/experience-bioluminescence/

https://www.nsf.gov/news/special_reports/science_nation/glowingsquid.jsp

Scuba diving in Hawaii: https://www.mantaraydiveshawaii.com/black-water-dive-ecoadventure/

Hawaiian diving: https://www.wsj.com/articles/diving-in-the-dark-off-hawaiis-coast-1412359144

Quorum sensing and infections:
https://www.sciencedaily.com/releases/2012/10/121011141435.htm
https://www.nature.com/news/2009/090121/full/news.2009.47.html

Glowing seafood
http://today.oregonstate.edu/archives/2010/aug/glow-dark-shrimp-don%E2%80%99t-fret-say-seafood-safety-experts%E2%80%A6

Glowing seafood in Oregon
https://www.opb.org/news/article/glowing-seafood-oregon-not-cause-concern/

Standards

Biology 1 Standards

LAFS.910.RST.1.1
Cite specific textual evidence to support analysis of science and technical texts, attending to the precise details of explanations or descriptions.
HE.912.C.1.5
Analyze strategies for prevention, detection, and treatment of communicable and chronic diseases.
LAFS.910.RST.1.2
Determine the central ideas or conclusions of a text; trace the text's explanation or depiction of a complex process, phenomenon, or concept; provide an accurate summary of the text.
LAFS.910.RST.1.3
Follow precisely a complex multistep procedure when carrying out experiments, taking measurements, or performing technical tasks, attending to special cases or exceptions defined in the text.
LAFS.910.RST.2.4
Determine the meaning of symbols, key terms, and other domain-specific words and phrases as they are used in a specific scientific or technical context relevant to grades 9–10 texts and topics.
LAFS.910.RST.3.7
Translate quantitative or technical information expressed in words in a text into visual form (e.g., a table or chart) and translate information expressed visually or mathematically (e.g., in an equation) into words.
SC.912.N.1.1

1. Define a problem based on a specific body of knowledge, for example: biology, chemistry, physics, and earth/space science, and do the following:
2. Pose questions about the natural world, (Articulate the purpose of the investigation and identify the relevant scientific concepts).
3. Conduct systematic observations, (Write procedures that are clear and replicable. Identify observables and examine relationships between test (independent) variable and outcome (dependent) variable. Employ appropriate methods for accurate and consistent observations; conduct and record measurements at appropriate levels of precision. Follow safety guidelines).
4. Examine books and other sources of information to see what is already known,
5. Review what is known in light of empirical evidence, (Examine whether available empirical evidence can be interpreted in terms of existing knowledge and models, and if not, modify or develop new models).
6. Plan investigations, (Design and evaluate a scientific investigation).
7. Use tools to gather, analyze, and interpret data (this includes the use of measurement in metric and other systems, and also the generation and

interpretation of graphical representations of data, including data tables and graphs), (Collect data or evidence in an organized way. Properly use instruments, equipment, and materials (e.g., scales, probeware, meter sticks, microscopes, computers) including set-up, calibration, technique, maintenance, and storage).

8. Pose answers, explanations, or descriptions of events,
9. Generate explanations that explicate or describe natural phenomena (inferences),
10. Use appropriate evidence and reasoning to justify these explanations to others,
11. Communicate results of scientific investigations, and
12. Evaluate the merits of the explanations produced by others.

SC.912.N.3.5
Describe the function of models in science, and identify the wide range of models used in science.
SC.912.L.14.3
Compare and contrast the general structures of plant and animal cells. Compare and contrast the general structures of prokaryotic and eukaryotic cells.
SC.912.L.14.4
Compare and contrast structure and function of various types of microscopes.
SC.912.L.15.15
Describe how mutation and genetic recombination increase genetic variation.
SC.912.L.15.4
Describe how and why organisms are hierarchically classified and based on evolutionary relationships.
SC.912.L.15.6
Discuss distinguishing characteristics of the domains and kingdoms of living organisms.
SC.912.L.16.3
Describe the basic process of DNA replication and how it relates to the transmission and conservation of the genetic information.
SC.912.L.16.5
Explain the basic processes of transcription and translation, and how they result in the expression of genes.
SC.912.L.18.11
Explain the role of enzymes as catalysts that lower the activation energy of biochemical reactions. Identify factors, such as pH and temperature, and their effect on enzyme activity.
SC.912.L.14.52
Explain the basic functions of the human immune system, including specific and nonspecific immune response, vaccines, and antibiotics.
SC.912.L.17.5
Analyze how population size is determined by births, deaths, immigration, emigration, and limiting factors (biotic and abiotic) that determine carrying capacity.
SC.912.L.16.10

Evaluate the impact of biotechnology on the individual, society and the environment, including medical and ethical issues.

Biology 1 Learning Goals

1. Students will be able design and evaluate scientific investigation using evidence of scientific thinking and problem solving.
2. Students will be able to interpret and analyze data to make predictions and defend conclusions.
3. Students will be able to use compound microscope to identify different cells and organisms.
4. Students will identify examples of scientific inferences made from observations.
5. Students will compare and/or contrast the structures found in prokaryotic cells and in eukaryotic cells.
6. Students will classify organisms based on the distinguishing characteristics of the domains and kingdoms of living organisms.
7. Student will be able to analyze cladograms and evaluate evolutionary relationship.
8. Students will explain the basic processes of transcription and/or translation and their roles in the expression of genes.
9. Students will explain how similarities in the genetic codes of organisms are due to common ancestry and the process of inheritance.
10. Students will evaluate examples and explain the possible impact of biotechnology on the environment.
11. Students will be able to measure rate of growth and use data about population dynamics, abiotic and biotic factors to explain a change in carrying capacity and its effect on population size in an ecosystem.
12. Students will identify and/or describe the effect of environmental factors on enzyme activity.

AP Biology

Big Idea 1: Learning objectives

The student is able to evaluate data-based evidence that describes evolutionary changes in the makeup of a population over time.

The student is able to connect evolutionary changes in a population over time to a change in the environment.

The student is able to create a phylogenetic tree or simple cladogram that correctly represent evolutionary history and speciation from a provided data set.

The student is able to use data from a real or simulated population(s), based on graphs or models of types of selection, to predict what will happen to the population in the future.

Big Idea 2: Learning goals
Organisms use feedback mechanisms to maintain their internal environment and respond to external environmental changes.

Learning objectives (may use operons)
The student can justify a claim made about the effects on a biological system at the molecular, physiological or organismal level when given a scenario in which one or more components within a negative regulatory system is altered.

The student is able to make predictions about how positive feedback mechanisms amplify activities and processes in organisms based on scientific theories and models.

The student is able to design a plan for collecting data to show that all biological systems (cells, organisms, populations, communities and ecosystems) are affected by complex biotic and abiotic interactions. Using cell density, biofilms, oxygen availability, etc

The student is able to analyze data to identify possible patterns and relationships between a biotic or abiotic factor and a biological system (cells, organisms, populations, communities or ecosystems). Using cell density, biofilms, oxygen availability, nutrient availability, symbiosis, etc

The student is able to connect concepts that describe mechanisms that regulate the timing and coordination of physiological events. Using quorum sensing

The student is able to connect concepts in and across domain(s) to predict how environmental factors affect responses to information and change behavior.

Big Idea 3: Learning goals
The student can predict how a change in a specific DNA or RNA sequence can result in changes in gene expression.

The student is able to explain how the regulation of gene expression is essential for the processes and structure that support efficient cell function.

The student is able to describe basic chemical processes for cell communication shared across evolutionary lines of descent.

The student is able to construct explanations of cell communication through cell-to-cell direct contact or through chemical signaling.

Lesson 1: Understanding the Relationship: Why work together

Objectives:

- Discuss night scuba diving sights in Hawaii (maybe Florida too, however luminous organism is different in Florida)
- Read up on V. fischeri and its symbionts (fish and Hawaiian squid). Explanation of the relationships and why they are beneficial.

Background:

Videos
https://www.nationalgeographic.com/travel/florida-land-and-sea/experience-bioluminescence/

Scuba diving in Hawaii: https://www.mantaraydiveshawaii.com/black-water-dive-ecoadventure/

Hawaiian diving: https://www.wsj.com/articles/diving-in-the-dark-off-hawaiis-coast-1412359144

Glowing seafood
http://today.oregonstate.edu/archives/2010/aug/glow-dark-shrimp-don%E2%80%99t-fret-say-seafood-safety-experts%E2%80%A6

Glowing seafood in Oregon
https://www.opb.org/news/article/glowing-seafood-oregon-not-cause-concern/

Quorum sensing and infections:
https://www.sciencedaily.com/releases/2012/10/121011141435.htm
https://www.nature.com/news/2009/090121/full/news.2009.47.html

Articles: TBD

Lesson 1 Standards

LAFS.910.RST.1.1
Cite specific textual evidence to support analysis of science and technical texts, attending to the precise details of explanations or descriptions.
LAFS.910.RST.1.2

Determine the central ideas or conclusions of a text; trace the text's explanation or depiction of a complex process, phenomenon, or concept; provide an accurate summary of the text.
LAFS.910.RST.1.3
Follow precisely a complex multistep procedure when carrying out experiments, taking measurements, or performing technical tasks, attending to special cases or exceptions defined in the text.
LAFS.910.RST.2.4
Determine the meaning of symbols, key terms, and other domain-specific words and phrases as they are used in a specific scientific or technical context relevant to grades 9–10 texts and topics.
LAFS.910.RST.3.7
Translate quantitative or technical information expressed in words in a text into visual form (e.g., a table or chart) and translate information expressed visually or mathematically (e.g., in an equation) into words.

Lesson 1 Student learning objectives

1. Student will be able to analyze the beneficial function of bacteria.
2. Student will be able to understand the importance of a specific type of symbiosis and its benefit to the symbionts.
3. Student will to determine the central and supporting ideas from a set of texts.
4. Students will identify examples of scientific inferences made from observations.
5. Student will be able to analyze data to determine how symbiosis works between the symbionts.
6. Student will be able to describe bioluminescence in Vibrio fischeri

AP Biology

7. The student is able to connect concepts that describe mechanisms that regulate the timing and coordination of physiological events. Using quorum sensing

8. The student is able to connect concepts in and across domain(s) to predict how environmental factors affect responses to information and change behavior.

Procedures:
Students will be assigned into groups of 3-4 individuals and each will be give an article to read and analyze. They will use the information to create a personal meaning map of their understanding of the interaction between the Hawaii bobtail squid and/or fish and Vibrio fischeri.

Assessment:
This project will be assessed based on how well the information in the articles is integrated to create an understandable mind map or personal meaning map with essential information.

Lesson 2: Culturing the bacteria

Objectives: Pipetting V. fischeri cells in growth broth into well plates; shaking overnight and observing the changes (bioluminescence and microscopy)

Lesson 2 Standards
SC.912.L.14.3
Compare and contrast the general structures of plant and animal cells. Compare and contrast the general structures of prokaryotic and eukaryotic cells.
SC.912.L.14.4
Compare and contrast structure and function of various types of microscopes.
SC.912.L.15.15
SC.912.N.1.1

1. Define a problem based on a specific body of knowledge, for example: biology, chemistry, physics, and earth/space science, and do the following:
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3. Conduct systematic observations, (Write procedures that are clear and replicable. Identify observables and examine relationships between test (independent) variable and outcome (dependent) variable. Employ appropriate methods for accurate and consistent observations; conduct and record measurements at appropriate levels of precision. Follow safety guidelines).
4. Examine books and other sources of information to see what is already known,
5. Review what is known in light of empirical evidence, (Examine whether available empirical evidence can be interpreted in terms of existing knowledge and models, and if not, modify or develop new models).
6. Plan investigations, (Design and evaluate a scientific investigation).
7. Use tools to gather, analyze, and interpret data (this includes the use of measurement in metric and other systems, and also the generation and interpretation of graphical representations of data, including data tables and graphs), (Collect data or evidence in an organized way. Properly use instruments, equipment, and materials (e.g., scales, probeware, meter

sticks, microscopes, computers) including set-up, calibration, technique, maintenance, and storage).
8. Pose answers, explanations, or descriptions of events,
9. Generate explanations that explicate or describe natural phenomena (inferences),
10. Use appropriate evidence and reasoning to justify these explanations to others,
11. Communicate results of scientific investigations, and
12. Evaluate the merits of the explanations produced by others.

Lesson 2 Learning Objectives

1. Student will identify the parts of a microscope.
2. Student will learn the use of micropipettes.
3. Student will culture Vibrio fischeri in growth broth and well plate.
4. Student will be able to prepare a wet mount and use the microscope to analyze the specimen.
5. Student will understand what is needed for bacteria growth.
6. Student will observe bacterial growth and bioluminescence.

Procedures:
Student will transfer some bacterial sample from a stock solution of Vibrio fischeri into Photobacterium broth and let it grow overnight. We mounts will be observed under the microscope.

The turbid culture will be aliquoted the next day into wells in a 96-well plate to form different patterns that spells out terms familiar to students (EHS, RAMS, LIFE, DNA, RNA, etc).

Template will be provided for pipetting.

Assessment:
This project will be assessed based on the growth of bacteria, presence of luminescence and proper formation of the patterns through the luminescence. Scientific explanation in lab notebook will also be assessed.

Lesson 3 How quickly they grow: Determining bacterial growth rate

Overview

Determining the growth rate of two strains of V. fischeri (fish and squid symbionts) using spectrophotometer.

- Student can use spectrophotometer (600nm) to measure optical density to determine rate of growth
- Student can analyzed data on optical density and luminescence of one of more strains of Vibrio fischeri
- Student can analyze data from another bacterial growth experiment

Lesson 3 Standards

SC.912.N.1.1

1. Define a problem based on a specific body of knowledge, for example: biology, chemistry, physics, and earth/space science, and do the following:
2. Pose questions about the natural world, (Articulate the purpose of the investigation and identify the relevant scientific concepts).
3. Conduct systematic observations, (Write procedures that are clear and replicable. Identify observables and examine relationships between test (independent) variable and outcome (dependent) variable. Employ appropriate methods for accurate and consistent observations; conduct and record measurements at appropriate levels of precision. Follow safety guidelines).
4. Examine books and other sources of information to see what is already known,
5. Review what is known in light of empirical evidence, (Examine whether available empirical evidence can be interpreted in terms of existing knowledge and models, and if not, modify or develop new models).
6. Plan investigations, (Design and evaluate a scientific investigation).
7. Use tools to gather, analyze, and interpret data (this includes the use of measurement in metric and other systems, and also the generation and interpretation of graphical representations of data, including data tables and graphs), (Collect data or evidence in an organized way. Properly use instruments, equipment, and materials (e.g., scales, probeware, meter sticks, microscopes, computers) including set-up, calibration, technique, maintenance, and storage).
8. Pose answers, explanations, or descriptions of events,
9. Generate explanations that explicate or describe natural phenomena (inferences),
10. Use appropriate evidence and reasoning to justify these explanations to others,
11. Communicate results of scientific investigations, and
12. Evaluate the merits of the explanations produced by others.

SC.912.N.3.5

Describe the function of models in science, and identify the wide range of models used in science.
SC.912.L.14.3
Compare and contrast the general structures of plant and animal cells. Compare and contrast the general structures of prokaryotic and eukaryotic cells.
SC.912.L.14.4
Compare and contrast structure and function of various types of microscopes.
SC.912.L.17.5
Analyze how population size is determined by births, deaths, immigration, emigration, and limiting factors (biotic and abiotic) that determine carrying capacity.

AP Biology
Big idea 1

1. The student is able to use data from a real or simulated population(s), based on graphs or models of types of selection, to predict what will happen to the population in the future.

Big idea 2

2. The student is able to design a plan for collecting data to show that all biological systems (cells, organisms, populations, communities and ecosystems) are affected by complex biotic and abiotic interactions. (Using cell density, biofilms, oxygen availability, etc)

3. The student is able to analyze data to identify possible patterns and relationships between a biotic or abiotic factor and a biological system (cells, organisms, populations, communities or ecosystems). (Using cell density, biofilms, oxygen availability, nutrient availability, symbiosis, etc)

4. Big idea 3: The student is able to construct explanations of cell communication through cell-to-cell direct contact or through chemical signaling.

Lesson 3 Learning Objectives

1. Student will be able to make dilutions.
2. Student will be able to use micropipettes to transfer solutions.
3. Student will understand how spectrophotometer works.
4. Students will learn to use spectrophotometer to measure solution turbidity as optical density to show bacterial growth.
5. Student will learn to use Microsoft Excel, perform statistical analysis (means, standard deviation, confidence interval and SEM), and plot appropriate graph for the data.
6. Student will analyze optical density data from this or other experiment using Microsoft Excel.

7. Student will propose additional research questions that could use this technique

Procedures

Student will make dilution of bacterial broth from lesson 2 and use it to measure optical density for bacterial growth. Since each period is 50 minutes long, measurement could be taken every 10 minutes to understand and observe the process, however data from another experiment will be analyze. To observe the different phases of the bacterial growth curve, measurement must be taken for a longer period, which will not be possible in a class or school day. Students have access to computer and use Excel or Google Sheet to calculate means, standard deviation and plot an appropriate graph with error bars.

Assessment:

Based on results of statistical analyses, organization of tables and graphs in lab notebook

Lesson 4 How does it work: Understanding Gene Regulation in Vibrio *fischeri*

Overview:
Understand gene regulation using Vibrio mutants.

Lesson 4 Standards
SC.912.L.16.3
Describe the basic process of DNA replication and how it relates to the transmission and conservation of the genetic information.
SC.912.L.16.5
Explain the basic processes of transcription and translation, and how they result in the expression of genes.
SC.912.L.18.11
Explain the role of enzymes as catalysts that lower the activation energy of biochemical reactions. Identify factors, such as pH and temperature, and their effect on enzyme activity.
SC.912.L.16.10
Evaluate the impact of biotechnology on the individual, society and the environment, including medical and ethical issues.

Lesson 5 Learning Objectives

1. Student will understand that living things communicate through different processes (hormones, cell-to-cell contacts, etc).
2. Student will be able to describe the process of quorum sensing in bacteria.
3. Student will be able to explain how bacteria communicate through quorum sensing and work together to produce luminescence.
4. Student will be able to sequence steps in the functioning of the operon regulating quorum sensing and bioluminescence in V. *fischeri.*
5. Student will be able analyze how changes in genetic makeup in V. *fischeri* mutants is responsible for changes in phenotypes.
6. Student will be able to describe transcription and translation.

AP Biology
Big Idea 2:

7. The student can justify a claim made about the effects on a biological system at the molecular, physiological or organismal level when given a scenario in which one or more components within a negative regulatory system is altered.

8. The student is able to make predictions about how positive feedback mechanisms amplify activities and processes in organisms based on scientific theories and models.

9. The student is able to connect concepts that describe mechanisms that regulate the timing and coordination of physiological events. (Using quorum sensing)

10. The student is able to connect concepts in and across domain(s) to predict how environmental factors affect responses to information and change behavior.

Big Idea 3:

11. The student can predict how a change in a specific DNA or RNA sequence can result in changes in gene expression.

12. The student is able to explain how the regulation of gene expression is essential for the processes and structure that support efficient cell function.

13. The student is able to describe basic chemical processes for cell communication shared across evolutionary lines of descent.

14. The student is able to construct explanations of cell communication through cell-to-cell direct contact or through chemical signaling.

Procedures:

Different mutants of V. fischeri will be cultured and the presence or absence of bioluminescence and any changes in growth patterns will be observed. These changes will be analyzed to determine how missing components (mutations) of the different operon have influenced the phenotype of the orgasnims.

Assessment:

Completion of a lab report that analyzes the experiment and provide a scientific explanation using CER.

Lesson 5: How are we related: Understanding evolutionary relationship of Vibrio family

Overview:
Bioinformatics: using BLAST to determine genetic homology between different species of Vibrio (infectious and noninfectious). Creating cladogram using these organisms.

Lesson 5 Standards
SC.912.L.15.4
Describe how and why organisms are hierarchically classified and based on evolutionary relationships.
SC.912.L.15.6
Discuss distinguishing characteristics of the domains and kingdoms of living organisms.
SC.912.L.16.3
Describe the basic process of DNA replication and how it relates to the transmission and conservation of the genetic information.

AP Biology
Big Idea 1

1. The student is able to evaluate data-based evidence that describes evolutionary changes in the makeup of a population over time.

2. The student is able to connect evolutionary changes in a population over time to a change in the environment.

3. The student is able to create a phylogenetic tree or simple cladogram that correctly represent evolutionary history and speciation from a provided data set.

Lesson 5 Learning Objectives

1. Students will classify organisms based on the distinguishing characteristics of the domains and kingdoms of living organisms.
2. Students will explain how similarities in the genetic codes of organisms are due to common ancestry and the process of inheritance.
3. Student will be able to use BLAST to obtain DNA and mRNA sequences of specific genes.
4. Student will be able to create, analyze cladograms and evaluate evolutionary relationship.

Background:
Infectious and noninfectious species of Vibrio
V. *alginolyticus*
V. *anguillarum*
V. *campbellii*
V. *casei*
V. coralliilyticus
V. *cholera*
V. *coralliilyticus*
V. *cyclitrophicus*
V. *diabolicus*
V. *fluvialis*
V. *furnissii*
V. *harveyi*
V. *lentus*
V. *mimicus*
V. *mytili*
V. *natriegens*
V. *ordalii*
V. *parahaemolyticus*
V. *pectenicida*
V. *tapetis*
V. *tubiashii*
V. *vulnificus*
V. fischeri changed to Aliivibrio fischeri (2007)

Aliivibrio genus
A. *finisterrensis*
A. fischeri (was V. *fischeri*)
A. l*ogei*
A. *salmonicida*
A. *sifiae*
A. *wodanis*

Some databases for bacterial genomic sequences

1. http://bacteria.ensembl.org/index.html

2. https://www.ncbi.nlm.nih.gov/genome/microbes/

3. Microbial Genome Database for Comparative Analysis: http://mbgd.genome.ad.jp/

4. JGI IMG Integrated Microbial Genomes & Microbiomes: https://img.jgi.doe.gov/

Procedures:
V. fischeri has been moved to a different genus. For this unit, the students could analyze genetic homology among species of the Vibrio genus or those of the Allivibrio genus. Adjustment will be made later. Students will be divided into groups of 3-4 and they will search for the DNA and/or mRNA sequence of one of the main genes in the bioluminescence operons.

Some operons and genes

- Lux operon is composed of different genes (luxCDABEG):
- Lux A and B codes for luciferase enzyme
- luxCDE codes for a fatty acid reductase complex that makes the fatty acids necessary for the luciferase mechanism.

- The operon is regulated by luxR and luxI
- luxR codes for a transcription factor
- luxi codes for an autoinducer

(AinR and AI-2 operons are also present I Vibrio fischeri and each is composed of multiple genes).

These genes are common to the bacteria in this genus and any of them could be used for evaluate evolutionary relationship. A specific one will be identify for the students to use.

The students have learned about these genes in earlier lessons (units). The students will initially learn about ancestral and derived traits and how to use them to create a cladogram. Students will obtain gene sequences for the different species and blast them to get the sequence divergence, which will be used to create cladograms.

Assessment:
The essential components of the cladograms will be assessed.