Amp It Up! Engineering/Technology and Industry Lesson Extension

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Course Name: Biology

Lesson/Unit Name: DNA and Biotechnology Applications

Science or Education Topic(s): DNA, Biotechnology, Science Procedure. Career and College exploration.

Engineering Technology Industry Related Field/Activity: DNA extraction and Sequencing Protocols

When Taught: Near end of year, or in second year advanced biology class (or biotech class)

Abstract:

This series of lessons is based on one of the aspects of biotechnology related to the work done at Thrive Bioscience. "Thrive Bioscience is an instrumentation company that aims to increase reproducibility and consistency to cell based experiments and cell culture. Founders and leadership of the company are an experienced team composed of industry leaders with successful track records for developing new lab instrumentation from concept through commercialization. Thrive has been working hard to understand the current problems facing modern science. One such example is contamination. Although contamination is a well-known obstacle in cell culture, it continues to slow down cutting edge, highly impactful science. Thrive Bioscience is currently designing and developing instrumentation that will significantly decrease contamination as well as address other emerging barriers in cell culture."

Three lab activities work for this industry extension lesson: Isolation of bacteria DNA (source: Cold Spring Harbor Laboratory), PCR Simulation Lab (source: University of Utah), and Investigating Mitosis (Source: Nuffield Foundation).

The last activity involves students working in teams to research regional biotechnology companies and create an informational poster on the company and their applications.

Objectives and assessment: Using the table below, identify at least 3-5 learning objectives (content and/or pedagogical) and describe how each will be assessed.

Objectives By the end of this lesson/unit, the students will be able to:	Assessment How was the objective assessed?.
Extract DNA from a bacteria sample Extract DNA from fruit Report on observations Document procedures	Lab report, observation
Identify PCR process: View PCR Simulation	Identify the process of PCR (Polymerase Chain

(possible link to Community College biotechnology program for lab activity at their campus)	Reaction).
Prepare and Complete a mitosis lab Document lab procedure Report on observations	Lab report, observation
Present information on one regional biotechnology company related to the lab and theory presented in the less.	Students will develop an informational poster on the company they research to present to the class.

Engineering/Technology Link:

1. How did you *introduce* engineering/ technology concepts or the company/industry focus in your course?

   - ✓ Defined terms (science, engineering, technology)
   - ✓ Described the scientific process/lab protocol
   - ✓ Overview of the company
   - ✓ Science related to the cell biology research field (DNA)

2. After introducing the concepts, what did/will the students do to explore and apply the engineering/technology and industry specific concepts?

Students will explore applications for each of the lab procedures completed in this lesson. Additionally, connections could be made by having the community college biotechnology faculty assist in a lab or offer a lab experience on their campus related to the DNA process. Finally, engaging the North Shore workforce investment board and partnering companies to explain job opportunities and applications of these lab procedures in the field would bring the lesson from academic application to college and career connections.

Level of Inquiry: Which of the following best describes the level of inquiry (adapted from Bell 2005) you used for this lesson/unit?

- ✓ *Structured inquiry*: Instructor provides question and procedure. Students determine the results based on given procedures.

Lesson Extension Plan:

Title/Topic: DNA structure and Application in the Biotechnology Industry

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Time (minutes): multiple classes depending on components selected, community college activities and WIB/business presentations.

Company Name and brief Description:
Thrive Bioscience is an instrumentation company that aims to increase reproducibility and consistency to cell based experiments and cell culture. Founders and leadership of the company are an experienced team composed of industry leaders with successful track records for developing new lab instrumentation from concept through commercialization.

Overview of the Lesson
This lesson involves introducing students to the field of biotechnology while learning about DNA, DNA production and mitosis. There are three possible lab activities as well as a career research activity

Standard(s)/Unit Goal(s) to be addressed in this lesson: (from the Next Generation Science Standards, 2015)

HS-LS1-1. Construct an explanation based on evidence for how the structure of DNA determines the structure of proteins which carry out the essential functions of life through systems of specialized cells. [*Assessment Boundary: Assessment does not include identification of specific cell or tissue types, whole body systems, specific protein structures and functions, or the biochemistry of protein synthesis.*]

HS-LS3-1. Ask questions to clarify relationships about the role of DNA and chromosomes in coding the instructions for characteristic traits passed from parents to offspring. [*Assessment Boundary: Assessment does not include the phases of meiosis or the biochemical mechanism of specific steps in the process.*]
HS-LS3-2. Make and defend a claim based on evidence that inheritable genetic variations may result from: (1) new genetic combinations through meiosis, (2) viable errors occurring during replication, and/or (3) mutations caused by environmental factors. [Clarification Statement: Emphasis is on using data to support arguments for the way variation occurs.] [Assessment Boundary: Assessment does not include the phases of meiosis or the biochemical mechanism of specific steps in the process.]
HS-LS1-4. Use a model to illustrate the role of cellular division (mitosis) and differentiation in producing and maintaining complex organisms. [*Assessment Boundary: Assessment does not include specific gene control mechanisms or rote memorization of the steps of mitosis.*]

Students will also engage in a number of scientific processes strengthening their ability to conduct research, communicate findings and make connections between academic practices and workplace applications in the biology and life science fields.

Essential Question(s) addressed in this lesson: Why do scientists extract DNA? How is DNA extracted from cells? How is DNA replicated? How can we replicate DNA in a lab? How does the ability to replicate DNA help us in the field?

Link to Industry:
Thrive has been working hard to understand the current problems facing science. One such example is contamination. Although contamination is a well-known obstacle in cell culture, it continues to slow down cutting edge, highly impactful science. Thrive Bioscience is currently designing and developing instrumentation that will significantly decrease contamination as well as address other emerging barriers in cell culture.

What students should know and be able to do before starting this lesson

Understand the importance of DNA and how carries out genetic blueprint.

Instructional Materials/Resources/Tools
Each lab has a list of materials needed to complete the labs. See links in lesson to those websites.
Appendix A: DNA Extraction Lab lists materials for that lab.

Lesson Delivery

Lesson Opening

Introduction to the company (above) . Then review of the topics covered by the industry area:
Mammalian cells that are commonly grown in petri dishes and flasks require at least a few specific environmental factors to maintain homeostasis. These includes a growth environment that contains 5% carbon dioxide, 95% humidity, and 37°C temperature. Additionally, cells must be grown in medium that allows provides appropriate nutrients for proper cell metabolism. Thrive aims to design and develop cell culture incubators that grow cells in optimal growth environment.

Basic elements of a cell's environment can have a significant impact on cells health and homeostasis. Therefore, it is of utmost importance for scientists to grow cells in consistent, ideal growth conditions. Experiments conducted on cells can become less reproducible when cells are not properly maintained. This type of scenario can be especially problematic in drug discovery companies that invest significant resources into long term projects unknowingly based on inconsistent, irreproducible data.

Cell culture/DNA concept

Many of the classic cell lines grown in culture are derived from tumors removed from patients. Cancer is undoubtably a devastating diagnosis and many people have spent their entire careers trying to find treatments and cures for this life threatening disease. However, cancer cells have been used as a tool by researchers for decades. One famous example are HeLa cells, which are cervical cancer cells. (Summer reading suggestion: "The immortal life of Henrietta Lacks", a book written about HeLa cells)

Researchers often grow a number of different cell lines from a number of different sources at one time. Each cell line is grown in its own flask and often has specific, unique media due to unique growth requirements (for example, cells from reproductive organs require hormones in media).

Despite best efforts, sometimes mistakes can happen. One mistake can be accidental mixing of 2 cell lines. HeLa cells grow well in a variety of environmental conditions, which means HeLa cells can often survive when unknowingly mixed with another cell line. Because these mistakes can happen without the researcher realizing, many companies routinely send their cell lines for verification testing. These tests involve extracting DNA from a group of cells, sequencing the isolated DNA, and matching the sequenced genome to a known source. In this way, the DNA extracted from cells acts like a fingerprint to allow researchers to identify and verify which cell lines they are working with in order to avoid conducting experiments with an incorrect cell line.

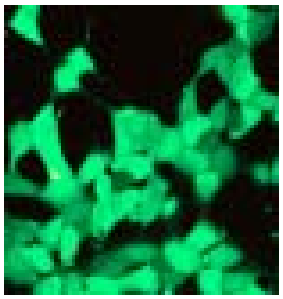
Organelle/Environment concept

The mitochondria is an organelle commonly referred to as the "powerhouse of the cell". This is because the mitochondria produces ATP, an important source of chemical energy for mammalian cells. The electron transport chain that produces ATP causes a positive gradient in the intermembrane space of the mitochondria, and a negative gradient in the mitochondrial matrix. This polarization of gradients is observed in healthy cells with active mitochondria.

When cells are under significant stress, it can cause decreased mitochondria activity. From a researcher's perspective, conducting experiments with cells that do not have active mitochondria may negatively affect experimental data. Therefore, researchers can choose to add a probe to their cells to better understand whether the mitochondria are polarized or depolarized.

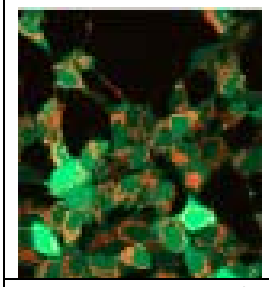
This probe, often called a mito tracker, passively diffuses across the cell membrane and the mitochondrial membrane. Once in the mitochondria, a moiety within the dye reacts with mitochondrial membrane proteins and is retained in polarized mitochondria. The mito tracker dye is fluorescent, allowing scientists to view their cells under a fluorescent microscope. If the dye is highly visible, the mitochondria are likely polarized/active.

If the cell is not healthy or is under stress caused by lack of nutrients, improper environmental conditions, etc., the mitochondria is often not active. In this scenario, addition of the fluorescent mito tracker probe will show almost no fluorescent staining in cells. This is because the integrity of the mitochondrial membranes are compromised, causing diffusion of the added probe.



Visualization of the mito tracker probe. This data has been taken and modified from the following published work: Gandhi S, Wood-Kaczmar A, Yao Z, et al. PINK1-Associated Parkinson's Disease Is Caused by Neuronal Vulnerability to Calcium-Induced Cell Death. *Molecular Cell*. 2009;33(5-3):627-638

Cells with active mitochondria that can take up the probe (there are many mitochondria in these cells, causing the entire cell to look fluorescent green):



Cells without highly active mitochondria that take up very little probe (red staining is separate from the mito tracker and can be ignored):

During the Lesson (activities/labs/challenges)

Students will then be guided through the following labs:

DNA Extraction Lab (Source: http://labcenter.dnalc.org/labs/dnaextraction/dnaextraction_d.html)
This resource provides a guided lab procedure to extract DNA from harmless e.coli (bacteria) using soap and ethanol. The resource provides background information, student lab manual, student protocol, result analysis and follow up activity linking activity to real world applications.

DNA Extraction Lab II (attachment I: created by Mary Kate Hartwell, Salem High School)

PCR Lab: The second lab (optional) involves engaging students in a simulated PCR (Polymerase Chain Reaction) lab offered by the University of Utah, Health Sciences department. This simulation allows the students to use the PCR tool to focus in on a segment of DNA and make billions of copies. This process is used in identifying bacteria and viruses. In the field, PCR could be used to diagnose illnesses or help in criminal investigations. (Source: Genetic Science Learning Center, University of Utah http://learn.genetics.utah.edu/content/labs/pcr/).

Lesson Closing

Finally, at the conclusion of the labs. Students working in teams will research regional biotechnology companies and develop a poster for one company in the region. Each poster will contain information the following information: Name of Company, Location, Types of Jobs posted (if any), Products or Services offered. Who their customer is, and a brief description of the company. Students may include photos (if available on the company webpage) and other interesting information.
If possible, a presentation by the Workforce Investment Board on labor markets in the biotechnology industry and related college degrees linked to the industry would be beneficial.

Assessment
Student Assessment: Student assessment will be based on the student lab journals, lab result write-ups and team poster presentations.
Delivery Assessment: Students should give feedback after each lab regarding level of understanding, engagement and relevancy to the field and the subject matter. Students can also make suggestions for improving the activities within this lesson for delivery modifications.

Attachment I: DNA Extraction Lab. Mary Kate Hartwell, Salem High School 2015

DNA Extraction Lab:

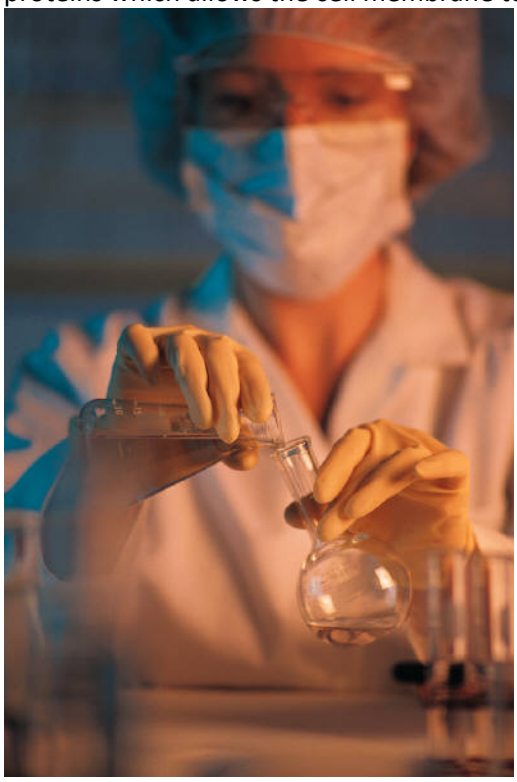
Purpose: To extract DNA from a Banana (plant cell). To understand the procedure and method of science used in this process.

Prior knowledge: Understand the importance of DNA and how carries out genetic blueprint.

The work being done a "Thrive" involves the extraction of Mitochondrial DNA. This lab will relate to a process.

The process of isolating DNA from a cell is the first step of many laboratory procedures in biotechnology. The Scientist must be able to separate DNA from the unwanted substances of the cell with out damaging the DNA.

A filtrate is made of bananas and treated with a buffer containing salt and distilled water. The salt solution will bind with the positive ions to prevent enzymes from chewing up the DNA. The salt will shield the negative phosphates of the DNA which allows these ends to come closer so that they can precipitate out of a cold alcohol solution. A detergent is added with causes the cell membrane to break down by emulsifying the fats and proteins which allows the cell membrane to break apart



Materials:

- 20 ml DNA extraction buffer (15g Salt and 1L water)
- 1ml liquid dishwashing detergent (Dawn)
- Ice cold isopropyl alcohol
- 1/3 banana (you can use spinach too, but need a blender)
- Ziplock bag
- Coffee filters
- Coffee stirrers
- 200ml beaker
- elastic
- test tube

Procedure

1. Put 1/3 of a banana into the ziplock bag, and squeeze out all the air.
2. Gently mash the banana to a pulp for about 2 minutes
3. Open the bag and add 20 ml of the extraction buffer to the bag
4. Filter the mixture through either cheese cloth or a coffee filter.
5. Save the liquid that is coming through
6. Pour 10 ml of the filtrate into a test tube
7. Add 1 ml of detergent. Mix gently with the coffee stirrer.
8. Immediately add 20 of ice cold alcohol to the test tube slowly pouring it down the side of the test tube to create a layer on top.
9. Let this sit fro about 2-3 minutes. Bubbles will form and DNA will precipitate out of the solution
10. Gently swirl the DNA with the coffee stirrer. It will look like a white mucus.
11. Take the DNA out and place on a paper towel and observe.