CSI Ann Arbor: A Middle School look at Polymerase Chain Reaction or PCR Technology Philip Lundy Scranton Middle School 8415 Maltby Road Brighton, MI 48116 plundy@bas.k12.mi.us Mentored by Dr. Lukacs, University of Michigan

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Teacher Section

Overview

Students will explore PCR (Polymerase Chain Reaction) technology and DNA fingerprinting via a student directed web quest Students will play the roll of a custodian working their way through college at a CSI (Crime Scene Investigation) lab. They will research technologies used in the lab to virtually perform a DNA fingerprint of a group of suspects implicated in the theft of the students food, including that staple of middle school diet, bubblegum. As an extension teachers will then have an option to present a lab in which the students will be able to extract DNA from plants using household materials.

Science Background

<u>PCR</u>

In the early eighties researchers were looking for ways to replicate small trace samples of DNA so that they may investigate the sample using conventional laboratory techniques. The solution to this problem came from an American Chemist named Kary Mullis. While driving down a deserted stretch of highway in 1983 he came up with an idea which would later win him the Nobel Prize. Kary Mullis had created the Polymerase Chain Reaction.

Trace samples of DNA are collected and placed into a solution of polymerases which serve to copy the DNA, primers which allow the copying to begin, and nucleotides which attach to the primers to copy the DNA strand.

This sample is then heated just below the boiling point to denature or split the two complimentary strands of the DNA. The temperature is then lowered to allow the primers to attach to the two DNA strands where the primers and the DNA match. The polymerases then begin to

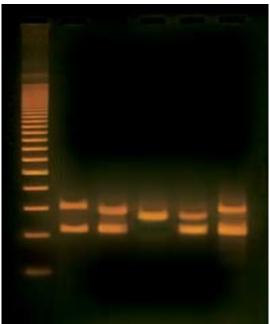
lengthen the growing DNA strands. The temperature is then raised again to a temperature which allows the polymerase to function at it's most efficient speed.

These steps are then repeated and after each cycle the number of DNA molecules is doubled. After 20 cycles a million copies of DNA are formed from a single piece of sample DNA.

DNA Fingerprinting

DNA fingerprinting got it's start in the United Kingdom the the early 1980's. This process was pioneered by Dr. Alex Jeffreys of the University of Leicester. The first criminal case featuring DNA Technologies took place in 1987.

After DNA is collected from a crime scene it is amplified using PCR technology. This sample, and all human DNA, contains polymorphic regions which vary in length from person to person. In this Web quest we used the Variable Number of Tandem Repeats (VNTR)



method of fingerprinting which focuses on an area of chromosome 1 which has a 16 nucleotide sequence which is repeated between 16 and 40 times. When this section of DNA is amplified the products are run through a electrophoresis gel which separates the samples by the length of the strands. When the gel is developed a black band or bands will form which can then be used to identify or clear a suspect. If the person is homozygous they will have equal numbers of repeats on each chromosome and their result will be a single band. More often then not the person will be heterozygous and their results will produce two distinct bands in the gel. When several different VNTR's are used a unique DNA fingerprint of an individual is produce which is unique to that person.

Edvotek PCR-based VNTR Human DNA Typing.

This web quest was based on the Edvotec Kit # 334. All protocols and background information used are from this kit which is available online at <u>www.edvotek.com</u>.

Learning Objectives

Students will review DNA basics Students will research PCR and DNA fingerprinting technology Students will complete a virtual lab procedure Students will apply their knowledge learned to produce a Power Point or movie presentation describing an invention of theirs using DNA fingerprinting technology.

Time Requirements

About 5 class periods are required for this web quest

Introduce and complete the web quest 2 - days. Research and produce student presentations 2 - days. Present student presentations 1 - day.

Advance Preparation

One sheet is recommended for each student and a team of two is recommended for the presentations. A guided notes sheet is available within the Web quest and on the Teachers page.

Materials and Equipment

For the web quest you will just need an Internet connection and a PC or Mac.

The CSI Ann Arbor website is located at <u>http://www.csiannarbor.homestead.com</u>

If you plan to do the DNA extraction as an extension you can use the website: <u>http://learn.genetics.utah.edu/units/activities/extraction/</u>

This protocol contains the clearest extraction technique out there.

The materials are as follows: 400 ml beakers

100 ml of a DNA source such as dried peas
1 ml of salt
200 ml of cold water
30 ml of detergent
Pinch of meat tenderizer
2 test tubes
blender
strainer

Student Prior Knowledge

Students should have completed their unit on DNA and heredity and have a basic understanding of how DNA replicates. The students should also have an understanding of phenotypes and genotypes and an understanding of heredity. This web quest was designed as an extension and was presented after the students finished the unit on DNA.

What is expected from students

The students should first complete their composition notes on DNA, PCR, and DNA Fingerprinting. These notes should be done individually while the students are collecting information.

The students will next produce a Power point presentation on their new business, career field, or invention to use the PCR and DNA fingerprinting technology they have discovered. This could be a completely new idea or an upgrade to an existing idea. The students should work in groups of two and plan on presenting their ideas to the class.

An student example is available at the link below and is included as a representation of an example of students work.

www.csiannarbor.homested.com/student_example.ppt

Assessment

A rubric is included in the web quest available at

http://www.csiannarbor.homestead.com/evaluationhtml

This basic rubric can be modified to your individual needs.

Name: _____

Hour: ____

Name three scientists whose work led to the discovery of the Structure of DNA.

What is DNA?

What is a DNA sequence?

What is a genome?

What is a chromosome?

What is a gene?

What is a karyotype?

What information can a karyotype give us?

What are the four bases of DNA?

What bases pair together?

Steps of DNA Replication:

1.)

2.)

3.)

What is a polymorphism?

What is a polymerase chain reaction (PCR) reaction?

Who invented the PCR reaction?

What is a PCR primer?

Define electrophoresis?

What is the electrophoresis gel?

How does electrophoresis work?

What is a DNA Fingerprint?

How are DNA fingerprints used to tell the difference between two individuals?

Draw a sketch of your gel results making note of the two DNA fingerprints that match.

Who has been eating all of your food and chewing your gum?

Key:

Name three scientists whose work led to the discovery of the Structure of DNA. Rosalind Franklin, James Watson, and Francis Crick.

What is DNA? Deoxyribonucleic acid. The instruction book for a cells activities.

What is a DNA sequence? The arrangement of bases in an organisms DNA.

What is a genome? The complete set of DNA in any organism.

What is a chromosome? Visible DNA.

What is a gene? A specific sequence of DNA which codes for a specific set of proteins. These proteins will determine a specific trait of an organism.

What is a karyotype? An organized profile of an organisms chromosomes.

What information can a karyotype give us? The sex of an organism or if some genetic abnormality exists.

What are the four bases of DNA? Adenine, guanine, cytosine, thymine.

What bases pair together? Adenine with thymine and cytosine with guanine.

Steps of DNA Replication:

- 1.) DNA molecule unzips at the connected bases.
- 2.) Each side of the DNA "Ladder" forms a template for the complementary side to form.
- 3.) Replication is complete with two identical strands of DNA.

What are nucleotide polymorphisms? Single letter changes in our DNA code effecting at least 1% of the population.

What is a polymerase chain reaction (PCR) reaction? A technique for quickly making many copies of a specific segment of DNA.

Who invented the PCR reaction? Kary Mullis

What is a PCR primer? A primer is a nucleic acid strand, or a related molecule that serves as a starting point for DNA replication.

Define electropheorisis? Separation of molecules (proteins or nucleic acids) in an electric field and through a gel as a function of their size or their electric charge.

What is the electropherosis gel? Electrophoresis Gel is a porous matrix, or meshwork, often made of carbohydrate chains with pores which serve to separate the different sizes of DNA strands.

How does electropheresis work? An electric field is generated to separate charged molecules that are suspended in a matrix or gel support. Negatively charged molecules move toward the anode, on one side of the gel, and positively charged molecules move toward the cathode, on the other side.

What is a DNA Fingerprint? The chemical/physical profile of an organism's nucleotide sequences, typically determined from segments of DNA that are 100 to 1,000 base pairs long. DNA fingerprints illuminate the genetic differences between and among individuals.

How are DNA fingerprints used to tell the difference between two individuals? Scientists use a small number of sequences of DNA that are known to vary among individuals a great deal, and analyze those to get a certain probability of a match.

Who has been eating all of your food and chewing your gum? Aaron.