Rationale
The polymerase chain reaction (PCR) is a fast and inexpensive technique used to amplify (copy) small segments of DNA. Because significant amounts of DNA are necessary for molecular and genetic analyses, studies of isolated pieces of DNA are nearly impossible without PCR amplification.

PCR is often heralded as one of the most important scientific advances in molecular biology. It revolutionized the study of DNA to such an extent that its creator, Kary B. Mullis, was awarded the Nobel Prize for Chemistry in 1993.

Objectives
Students will:

- List and describe the three steps used in Polymerase Chain Reaction and explain how repeating these three steps amplify DNA exponentially.
- Perform a PCR reaction.
- Describe how an agarose gel separates DNA fragments according to size.
- Explain how differences in DNA sequence result in different patterns of bands on a DNA electrophoresis gel.
- Analyze banding patterns for similarities and differences among individuals.
Key Points
I. See Power Point for Lesson 2 Objective 2.2

Engage
- Show students a real world application using VNTRs and PCR by visiting this website:
  Ask students how PCR could be helpful in crime scene investigations.

Activity
1. Pre-Lab Activities: Several pre-lab activities are listed below. All of the activities will introduce students to PCR. Choose one or all of them based on time and the level of your students.
   - PCR Cycle Sketch
   - [PCR Virtual Lab](http://www.teachersdomain.org/resource/tdc02_sci_life_gen_forensics/)
   - [PCR Game](http://www.teachersdomain.org/resource/tdc02_sci_life_gen_forensics/): Have students complete and print off the final screen to turn in.

2. Lab Activity: There are several PCR biotechnology kits offered through your district approved science supply vendor. Kits that are developed around a crime scene investigation are usually very engaging for students and allows for future studies in forensic science. Most kits come with extensive teacher intrusions, student protocols and lesson plan ideas.
   a. Students can set up a crime scene and use PCR technology to identify suspects.
   b. A grading rubric has been provided as a sample crime scene scenario.

3. Application: Case #1 at [http://forensics.rice.edu/](http://forensics.rice.edu/) refers to applications of PCR using CODIS. Visit the “Fun Stuff” tab for an educators guide.

4. Extension: Have students research Real-Time PCR vs. traditional PCR.

Materials
- [Worksheet: “PCR Virtual Lab”](http://www.teachersdomain.org/resource/tdc02_sci_life_gen_forensics/)
- [Worksheet: “PCR Cycle Sketch”](http://www.teachersdomain.org/resource/tdc02_sci_life_gen_forensics/)
- [PCR Game](http://www.teachersdomain.org/resource/tdc02_sci_life_gen_forensics/)
- PCR Crime Scene Grading Criteria
Assessment
- Student answer pre-lab activity worksheets correctly
- PCR Crime Scene Grading Criteria

Accommodations for Learning Differences
- Visit the Special Populations section of the CTE Career and Technical Education Website: http://cte.unt.edu/special-pops.

National and State Education Standards

**Texas College and Career Readiness Standards**

I. Nature of Science: Scientific Ways of Learning and Thinking
   - C1, C2, C3, D1, D2, D3, E1, E2

II. Foundation Skills: Scientific Applications of Mathematics
    - A2, A5, A7

III. Foundation Skills: Scientific Applications of Communication
     - A1, B1, B2, B3, D1

IV. A1, E1
PCR Cycle Sketch

For this assignment you will be drawing several cycles of PCR and answering a few short questions. The following animations will help you to understand the processes of PCR and gel electrophoresis.

- [http://www.dnalc.org/ddnalc/resources/pcr.html](http://www.dnalc.org/ddnalc/resources/pcr.html)

1. A thorough understanding of the process of PCR will help in the analysis of your DNA gel. The figure below represents a segment of double-stranded DNA with 100 basepair segments denoted by each letter. The primers are indicated by the arrows for both the sense and anti-sense DNA strands. In the following questions, a “copy” of DNA refers to a double-stranded piece of DNA.

   ![DNA sketch](Image)

   A. Draw the first three cycles of PCR indicating the
      a. 2pts: intermediate products (products that are produced before the targeted sequence) labeled with letter designations (label blue)
      b. 2pts: directionality (5′ → 3′), (label red)
      c. 2pts: TAQ polymerase (label purple)
      d. 2pts: size of the desired target double-stranded DNA product (label green)
      e. 7pts: include the temperatures (label orange) and their significance (pen/black) for each step in the PCR reaction for the first cycle

Copyright © Texas Education Agency 2012. All rights reserved.
2. Answer the following questions assuming there have been four cycles of PCR. (Hint: Look for graph analysis using one of the above links!)

A. What is the total number of target DNA copies and the total number of intermediate DNA copies assuming you began with one double strand DNA template? Note: Target DNA copies contain only the target DNA on both strands.

Target DNA copies 2pts __________ Intermediate DNA copies__________ 2pts

B. What is the ratio of intermediate to target DNA copies in the fourth cycle? How would this ratio change after twenty cycles? How does this ratio affect what you see in the DNA gel? 3pts

C. Is there an equation that can be used to predict copy number? What is it? 3pts
PCR Crime Scene Rubric

Name: ____________________

In the Laboratory:

_____ (5 points) Student is proficient in using the micropipette (40ul in PCR tube).

_____ (5 points) Student is proficient at making an agarose gel.

_____ (5 points) Student is proficient at loading samples under buffer.

Analysis of Results:

_____ (5 points) Data collected in table.

_____ (10 points) Questions are answered completely and with no errors.

Application:

_____ (10 points) Go to http://forensics.rice.edu/. Complete case #1. Print off certificate once completed and attached to this rubric.

Extension:

_____ (5 points) Research the difference between Real Time PCR and traditional PCR. List 3 similarities and 3 differences on the back of this rubric.

Lab Station Clean and Set Up Correctly

_____ (5 points)