

**Appendix A. Phillips *et al.***  
**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.

The Polymerase Chain Reaction by Sumanas, Inc.:

<http://www.sumanasinc.com/webcontent/anisamples/molecularbiology/pcr.html>

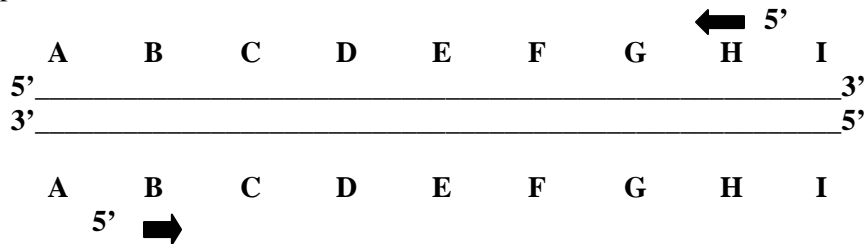
Polymerase Chain Reaction by the Dolan DNA Learning Center:

<http://www.dnalc.org/ddnalc/resources/pcr.html>

Gel Electrophoresis by the Dolan DNA Learning Center:

<http://www.dnalc.org/ddnalc/resources/electrophoresis.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 base pair 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.



- A. Draw the first three cycles of PCR indicating the intermediate products labeled with letter designations, directionality ( $5' \rightarrow 3'$ ), and the size of the desired target double-stranded DNA product. Include the temperatures and their significance for each step in the PCR reaction for the first cycle.
- B. For each cycle, indicate how many copies of target double stranded-DNA and “intermediate DNA” (the DNA includes target DNA region plus a bit of the flanking DNA and/or the original DNA strand) you would have after each cycle assuming you began with one double strand DNA template.
- 2) Answer the following questions assuming there have been four cycles of PCR.
- 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 \_\_\_\_\_ Intermediate DNA copies \_\_\_\_\_

- 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?