BIO 184 - PAL Problem Set Lecture 2 (Brooker Chapter 11)
DNA Replication

Section A. Experiments elucidating the mechanisms of DNA replication

How did Watson and Crick's double helix model suggest a mechanism for DNA replication?

Define the following DNA replication terms:

Template strand

Daughter strand

Parental strand

What does it mean that DNA replication is semi-conservative?

What three DNA replication models were tested by the Meselson and Stahl experiment?

How did Meselson and Stahl distinguish between previously replicated DNA and newly replicated DNA? How does this relate to parental strands and daughter strands?

What is the difference between light, half-heavy, and heavy DNA?

What was the purpose of a cesium chloride gradient?

What do each of the three DNA replication models predict in the Meselson and Stahl experiment?

The first generation of replication in the Meselson and Stahl experiment disproved which theory of replication?

In the Meselson and Stahl experiment, what happened to the heavy DNA strands by the third generation?

If Meselson and Stahl had found that DNA is replicated in a *conservative* manner, what results would these scientists see on the CsCl gradient after growing the organism for 1 generation?

Why was the Meselson and Stahl experiment so important? What was its conclusion?

Section B. DNA replication in bacteria.

Where on chromosomes does DNA synthesis initiate?

What types of DNA sequences are located in this region?

What are DnaA boxes? Where are they located?

What is the significance of the AT-rich region near the *ori*?

In which direction does DNA replication proceed?

How is RNA involved in DNA replication *in vivo*?

What does it mean to "prime" DNA replication?

Does DNA replication always need to be primed? Why or why not?

Does RNA synthesis need to be primed? Why or why not?

What is the name of the enzyme with RNA Polymerase activity that is directly involved in DNA replication?

What is DNA helicase? What would be inhibited during DNA replication if the gene encoding DNA helicase was mutated?

How is supercoiling introduced during DNA replication? How is supercoiling subsequently relieved?

What is DNA gyrase? What would be inhibited during DNA replication if the gene encoding DNA helicase was mutated?

Which enzyme fills in small regions of DNA where the RNA primers were located?

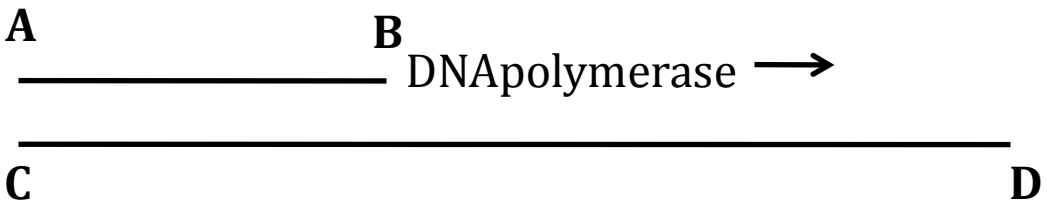
How many DNA polymerases does *E. coli* have? What is the difference between the different polymerases?

Which polymerase synthesizes the majority of DNA in the lagging strand?

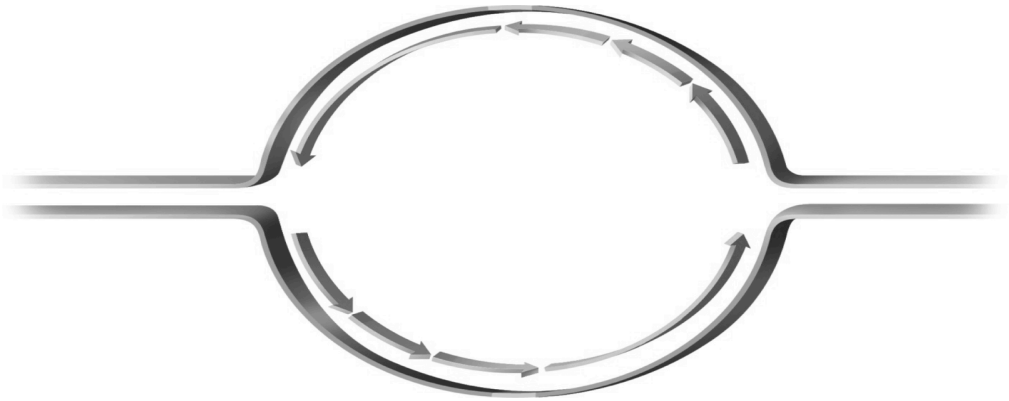
What does it mean if DNA replication has "high fidelity" or "low fidelity"?

What does it mean when a DNA polymerase is highly processive?

The diagram below represents two complementary strands of DNA. The one on the bottom is a template strand, and the one on the top is being synthesized by DNA polymerase in the direction shown by the arrow. Label A, B, C, and D appropriately with either 5' or 3' ends.



Below are two replication forks heading in opposite directions away from the *ori*. Label each strand appropriately with the following terms: parental strand, daughter strand, leading strand, leading strand template, lagging strand, lagging strand template. Also, label the 3' and 5' ends of each strand.



What two types of bonds form each time a new nucleotide is added to a growing DNA strand?

In what direction does DNA polymerase add nucleotides to a growing daughter strand? What direction does it read along the template?

What does it mean by the statement "DNA replication is bidirection"?

What does it mean that DNA replication is semi-discontinuous?

Are lagging strands synthesized continuously or discontinuously? Leading strands?

What are Okazaki fragments? How are Okazaki fragments ligated together?

If DNA polymerase could perform DNA synthesis in both directions (5'-3' and 3'-5'), then what would not be needed?

What sequence(s) are involved in bacterial DNA replication termination?

How does DNA Polymerase III proofread?

What secondary activity by DNA Polymerase III is responsible for proofreading?

What is the difference between an exonuclease and endonuclease?

Section C. DNA replication in eukaryotes.

What DNA replication problems are encountered with circular chromosomes?

What are catenanes? How are they removed?

What DNA replication problems are encountered with linear chromosomes?

What is the end-replication problem?

What are telomeres? Where are they located? What features do they have within their DNA sequence?

Do mitochondrial DNA have telomeres? Why or why not?

When is telomerase active? When is it not active?

Section D. PCR

List the 5 ingredients in a PCR reaction

List the 3 repeated steps of PCR. List the temperature for each step. What occurs during each step?

Step	Temperature	What occurs?

How does PCR target a specific genomic region of interest?

Why does PCR use Taq Polymerase and not other DNA polymerases, such as *E. coli*'s DNA polymerase?

What determines the exact size of a PCR product?

How does PCR compare to DNA replication *in vivo*? Fill out the table below.

Activity	Cellular replication	PCR
Denature DNA		
Primers		
Extension		
Number of copies produced		
Size of region copied		
Ingredients		
Purpose		

If a PCR reaction is seeded with a single DNA molecule, how many copies of the target sequence will be present after 22 cycles? (Assume that the reaction is 100% efficient)