

## 7.013 Recitation 6 - 2013

### Summary of Lectures 10 & 11:

**Replication:** The process of DNA replication occurs when two double-stranded DNA molecules are made from one double-stranded DNA molecule. This process occurs only in actively dividing cells because DNA replication always precedes cell division. The enzyme that catalyzes DNA replication is the protein DNA polymerase. DNA polymerase catalyzes the reaction of forming a phosphodiester bond between two deoxyribonucleotides. The start signal for DNA polymerase is an origin of replication, which is a site on DNA that may or may not be inside a gene. DNA polymerase proceeds down a piece of DNA until the entire genome is replicated. The new strands are synthesized in the 5' to 3' direction.

DNA replication occurs in a semi-conservative fashion. This means that, when one double-stranded molecule of DNA is replicated, the original double-stranded molecule is unraveled such that it is two separate "old" strands. Then each "old" strand acts as a template for one "new" strand. At the end of replication, each of the two new double-stranded molecules consists of one "old" strand running antiparallel to one "new" strand.

Besides DNA polymerase, two other enzymes play critical roles in DNA replication. Helicase unwinds the two strands of DNA such that they can be replicated. DNA ligase catalyzes the formation of a covalent bond between two adjacent nucleotides, and is used to seal the gap between the replicated fragments of DNA that are produced on the lagging strands of a replication fork. The leading strand is the strand that is being replicated in the same direction that the fork is being unraveled by helicase; thus DNA polymerase can just replicate this strand as one long continuous piece. The lagging strand is being replicated in the opposite direction that the fork is being unraveled, so DNA polymerase has to constantly hop on, replicate a piece, and then fall off and hop back on again. This leads to the strand being replicated in pieces that must be joined together by DNA ligase.

Mutations in DNA can be repaired by the proofreading mechanism of DNA polymerase (this works during replication), mismatch repair and excision repair mechanisms.

**Transcription:** The process of transcription occurs when a double-stranded DNA is unwound and one strand is transcribed to make RNA. The RNA that is transcribed from a gene can be one of three types of RNA: mRNA, tRNA or rRNA. All of these types of RNA are identical in their chemical composition, but they differ in their function. mRNAs are transcribed from genes that encode proteins; these mRNAs will go on to be translated by ribosomes. The rRNAs and tRNAs are transcribed directly from their respective genes, but they are never translated. Instead, they participate in the process of translating mRNAs. The rRNAs complex with ribosomal proteins to form the ribosome. Each tRNA becomes covalently attached to the correct amino acid, and then donates that amino acid to the ribosome whenever that amino acid is needed for translation of a protein.

### Questions

1. Say you wanted to replicate the following piece of DNA:

5' -GTACGTTTACGCCGTATATATCGTCGTAATGCTACGTAGCTCTACGAACA-3'  
3' -CATGCAAATGCGGCATATATAGCAGCATTACGATGCATCGAGATGCTTGT-5'

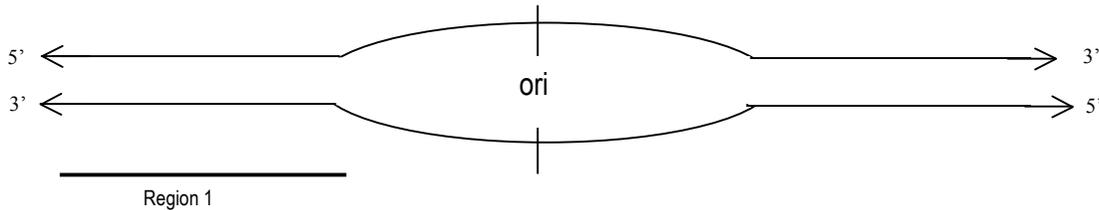
a) Design a 10-bp-long primer you would use to generate a new copy of the entire bottom strand of DNA. Label the 5' and 3' ends of the primer.

b) In what direction does the **new strand** of DNA get synthesized?

c) In what direction is the **template strand** for DNA synthesis read?

d) In the cell during DNA replication, the cell is not heated up each time to denature the original duplex. How does the cell separate the two strands of its double-stranded DNA during DNA replication?

2. Consider the following segment of the DNA that is a part of a much larger molecule constituting a chromosome. The sequence of region 1 is shown below.



Region 1:                    5' ...ATTCGTACGATCGACTGACTGACAGTC...3'   
                                   3' ...TAAGCATGCTAGCTGACTGACTGTCAG...5'

a) If we assume that a fragment of the lagging strand is made from region 1, what will be its sequence? Label the 5' and the 3' ends.

b) Why is DNA synthesis continuous at one strand and discontinuous at the other strand?

c) State the three major processes of DNA repair, which you were introduced to in 7.013 lectures.

3. The following is a partial sequence from the hypothetical gene, gene X. The boxed region is the promoter, and the direction of transcription is indicated by the arrow. Transcription begins at and includes the first G/C base pair after the box.

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5' ATCAGACAACGTCTCATGGGAGTACTTGGATGGAAGAGTAGAAGGTCATGACCAACCTCTTCCAATCCAACCACAAACAG
+-----+-----+-----+-----+-----+-----+-----+-----+-----+-----+-----+
3' TAGTCTGTTGCAGAGTACCCTCATGAACCTACCTTCATCTTCAGTACTGGTTGGAGAAGGTTAGGTTGGTGTGTTGTC

5' AAAATCAGCCAATATGTCCGACTTCGAGAACAAGAACCCCAACAACGTCCTTGGCGGACACAAGGCCACCCTTCACAACC
+-----+-----+-----+-----+-----+-----+-----+-----+-----+-----+-----+
3' TTTTAGTCGGTTATACAGGCTGAAGCTCTTGTCTTGGGGTTGTTGCAGGAACCGCCTGTGTTCCGGTGGGAAGTGTGG
  
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What are the first 10 nucleotides of the mRNA produced from gene X?

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