DNA Structure and Function

Deoxyribonucleic acid, DNA for short, is often referred to colloquially as "the stuff of life". What does that mean? It means that cells build bodies according to instructions encoded in nucleotide sequences of DNA. Specifically, those instructions specify the sequence of amino acids in each polypeptide that makes up a protein. In short, DNA codes for proteins and proteins are in turn involved in virtually every metabolic pathway involving all classes of biological molecules, often serving as enzymes that catalyze each reaction in a pathway. Another simplified way to put it is to say that DNA tells the cell what to do.

If DNA controls the activities of cells, then it follows that before a cell can divide, its DNA must be replicated so that each daughter cell receives an identical copy of the instructions. Indeed the continuity of life itself hinges on DNA's ability to replicate.

Thus we can say that DNA performs two critical functions:

- 1. It replicates
- 2. It serves as a master copy of instructions for making proteins

In this lab, you will model these two functions. For both procedures, you will be using a plastic model kit that contains the various players of DNA replication, transcription, and translation. The following is a quick guide to what the various pieces represent. Take a few moments to identify and organize the pieces in the kit. Refer back to this guide as needed.

Pre-assembled DNA and RNA nucleotides:

- Phosphate group: white tube
- Deoxyribose: black connector
- Ribose: purple connector
- Cytosine: blue tube
- Adenine: orange tube
- Guanine: yellow tube
- Thymine (DNA): green tube
- Uracil (RNA): purple tube



Transcription and translation pieces:

- Amino acids: black sticks. These fit onto one side of the tRNA molecule
- **tRNA molecules**: purple sticks. Note that you will have to place appropriate-colored tubes onto these to represent the anti-codon
- Peptide bonds: grey tube
- Ribosomes: 2-part manila paper



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Procedure 1: Modeling DNA replication

In this procedure, you model DNA replication. First, you build a parental DNA segment then, you replicate it. In this exercise, your hands act like the enzymes involved in the process.

Materials

Pre-assembled DNA and RNA nucleotides

Method

Making a parental DNA segment

- Connect DNA nucleotides to make up a single DNA segment that is 10 nucleotides long. The order of the nucleotides is unimportant; just be sure to include some of all 4 kinds in your segment.
- 2. Using base-pairing rules, and adding to the 3' end of the molecule, make the strand that is complimentary to the one you just made.
- 3. Connect the two strands using the small white connectors, which represent hydrogen bonds that connect complimentary bases.
- 4. Make sure your strands are antiparallel. Show your instructor your segment of DNA.

Modeling the replication of the DNA segment

- 1. Create a replication fork by pulling 4-5 nucleotide bases apart.
- 2. Create two new strands of DNA that are complimentary to the parental strands, continuing to separate the parental DNA strands as needed. Ensure that the newly created strands are antiparallel to the parental strands.
- 3. Verify that you have two identical segments of DNA by superimposing them: the bases should all match. Show your instructor your duplicated DNA segments.

Procedure 2: Modeling transcription and translation

Cells only decode one strand of the double-stranded DNA molecule: a gene consists of a single, specific and unique sequence of nucleotides. In this procedure therefore, you will use only one strand of your DNA molecule. Use your textbook or online resources to guide you through the process.

Materials

One strand of the DNA model from procedure 1 Ribosome tRNA molecules with appropriate anticodon bases Pre-assembled RNA nucleotides Amino acids

Method

- 1. With your lab partners, decide which strand of DNA you will transcribe, unzip the two strands, and set the unused strand aside.
- 2. Model the actions of **RNA polymerase** by adding RNA nucleotides complimentary to the DNA template strand. As in DNA replication, ensure the RNA transcript is antiparallel to the DNA template strand and that you are adding nucleotides only to the 3' end of the growing transcript.
- 3. As the RNA transcript grows, peel its completed end away from the DNA template to allow the complimentary strands of DNA to come back together.
- 4. The completed RNA transcript should have the same sequence of nucleotides as the nontemplate DNA strand, except that the RNA strand will have a Uracil wherever a Thymine appears in the DNA. Your mRNA transcript is ready for translation.
- 5. Set the completed mRNA transcript and atop the small subunit of the paper ribosome with its 5' end facing to the left.
- 6. Prepare tRNA models by placing 3 nitrogenous bases on the anticodon side of the purple model complimentary to the first three nucleotides (=the first codon) on the mRNA. To the other side of the tRNA model, attach an amino acid model (black stick). Use the genetic code table below to determine exactly which amino acid your model is intended to represent. Write it down here: _______.Remember that the codon is given in the language of mRNA, therefore, you will have to look a the triplet of nucleotides on the mRNA to determine the identity of the amino acid.
- 7. Match the anticodon on the tRNA to the codon on the mRNA strand. Slip the large subunit of the ribosome underneath the complex such that the tRNA is in the ribosome's P-site.
- 8. Prepare the next tRNA model in the sequence as you did in step 6. Which amino acid does the mRNA prescribe for this second codon?______
- 9. Match the anticodon on the tRNA to the codon on the mRNA strand. This second tRNA molecule should be in the A-site of the ribosome.
- 10. Model peptide bond formation by connecting the two amino acids in the ribosome with a grey peptide bond.
- 11. Release the amino acid from the first tRNA molecule, then model translocation by scooting the mRNA/tRNA complex three nucleotides to the left. The first tRNA molecule, now free of its amino acid, should be in the E-site of the ribosome. It is ready to exit the ribosome to go and be charged with a new amino acid.
- 12. Prepare the third tRNA model in the sequence as you did in steps 6 and 8. Which amino acid does the mRNA prescribe for this third codon?_____
- 13. Repeat steps 9, 10, and 11. You should now have a short polypeptide consisting of three amino acids. If you had a strand of DNA 300 nucleotides long, you could model the building of a polypeptide that is 100 amino acids long.

Congratulations! You are on your way to making nothing less than a whole organism, one protein at a time.

Figure 10.11				
Second base of RNA codon				
U	С	Α	G	
UUU UUC_ (Phenylalanin UUC_ (Phe)	ucu ucc Serine	UAU UAC(Tyr)	UGU Cysteine UGC (Cys)	U C
	UCA (Ser)	UAA Stop	UGA Stop	A
CUU CUC CUC CUC CUC CUA (Leu)	CCU CCC CCA (Proline (Pro)	CAU Histidine CAC (His)	CGU CGC CGA CGA CGA	Codon
		CAG_ (GIn)	CGG_	e RNA
AUU AUC AUC AUL AUC (Ile)	ACU ACC	AAU Asparagine AAC (Asn)	AGU Serine AGC (Ser) AGA Arginine	V O C Third base o
AUG Met or start	ACG_	AAG_ (Lys)	AGG_ (Arg)	G
GUU GUC GUA (Val) GUG	GCU GCCAlanine GCA (Ala) GCG_	GAUAspartic GACacid (Asp) GAAGlutamic GAGacid (Glu)	GGU GGC <u>Glycine</u> GGA (Gly) GGG_	U C A G