

Biology 1305
Modern Concepts in Bioscience (ICB Textbook)

Hello and welcome to the weekly resources for BIO-1305 - Biology 1

This week is Week 5 of class, and typically in this week of the semester, your professors are covering the topics below. If you do not see the topics your particular section of class is learning this week, please take a look at other weekly resources listed on our website for additional topics throughout the semester.

We also invite you to **look at the group tutoring chart on our website to see if this course has a group tutoring session offered this semester.**

If you have any questions about these study guides, group tutoring sessions, private 30 minute tutoring appointments, the Baylor Tutoring YouTube channel, or any tutoring services we offer, please visit our website www.baylor.edu/tutoring or call our drop in center during open business hours (M-Th 9am-8pm on class days at 254-710-4135).

KEYWORDS: Translation, tRNA, codons, mRNA processing, peptide bonds

TOPIC OF THE WEEK

Translation

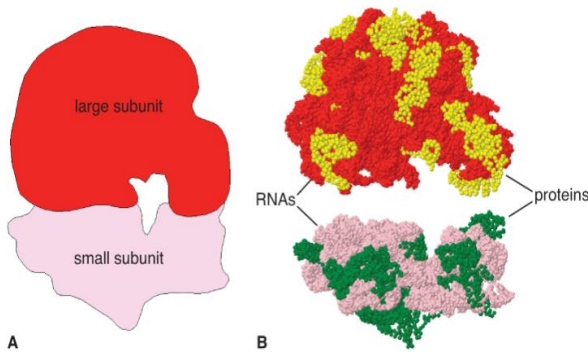
To recap, last week we looked at transcription and went over some of the experiments that investigators conducted to determine the functions of mRNA, tRNA, and rRNA. This week, we will focus on the creation of a polypeptide from mRNA codons, which are encoded by the original DNA nucleotide sequence.

Remember: The Central Dogma describes the process by which the information in DNA is passed on through RNA (**transcription**), which is then used to create a functional protein product (**translation**).

Translation: The process of converting RNA to protein. This takes place in the cytosol in ribosomes and requires **tRNA** to transfer amino acids to the growing polypeptide chain.

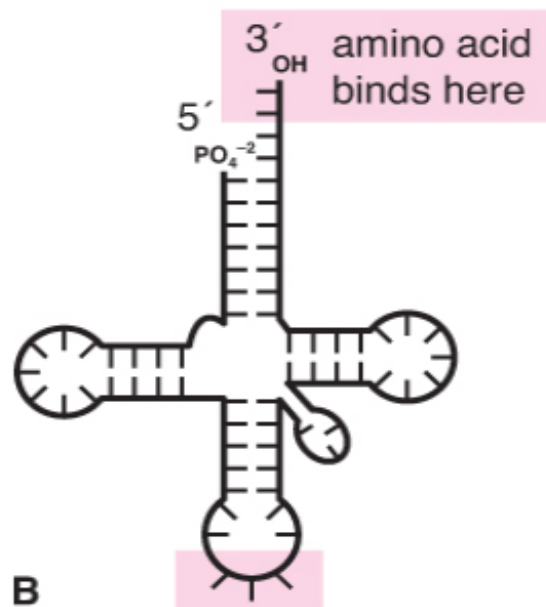
Heyer and Paradise, unless otherwise specified.

First, let's look at some of the key components of translation!



Recall from section 2.1 that the ribosome is composed of two subunits, both of which are made up of **protein and rRNA**. This helps us understand why **rRNA is the most abundant form of RNA** and makes up 80% of all RNA in a cell.

With the help of an enzyme, **ATP** energy is used to **covalently** link the correct amino acid to one end of the tRNA molecule. The tRNA molecule also has an **anticodon** at the other end, which attaches to an mRNA **codon** and ensures that the amino acids are added to the growing **polypeptide** in the correct place.



A ribosome moves along the mRNA strand, and at each codon, it “reads” the mRNA sequence to determine the correct amino acid, which is then brought to the ribosome by tRNA. This process of a ribosome moving along an mRNA strand consumes energy in the form of **GTP** (an energy-rich molecule which is similar to ATP, but with a guanine nucleotide instead of adenine).

(Remember: mRNA is encoded by the genetic information in the DNA and moves from the nucleus to the ribosomes in the cytosol.)

A group of researchers decided to attempt to reproduce translation “in vitro,” or in a test tube in the lab, to see how well they understood how the process works in cells. In order to do this, they assembled **a buffer at pH 7.2, chromosomal DNA, sources of energy in the form of**

adenosine triphosphate (ATP) and guanosine triphosphate (GTP), tRNAs, ribosomes, mRNA, and radioactive amino acids. The radioactive amino acids would allow them to visualize any proteins produced *in vitro*. In order to understand which of these components were required for translation and which weren't, they created seven different test tubes with different combinations of molecules; one was a control and six were experimental.

- *Where have we seen this process of radioactive labeling of amino acids for visualization in previous sections?*
- *Why do you think they chose pH 7.2 for the buffer?*

+		+	+	+	+	+	chromosomes
+	+		+	+	+	+	energy (ATP and GTP)
+	+	+		+	+	+	tRNA
+	+	+	+		+	+	ribosomes (65% rRNA, 35% protein)
+	+	+	+	+		+	mRNAs
+	+	+	+	+	+		amino acids

Positive Control: Column 1

--This positive control test tube contained all six components.

For the remaining test tubes, they omitted a different ingredient in each test tube, then loaded the mixtures onto a gel and used x-ray visualization to see if any large proteins had been produced. For example, the test tube that is represented by column 2 contained everything except for chromosomes.

Note: The individual amino acids that they added were very small and migrated out of the bottom of the gel; they did not appear on the x-ray film. The gel only showed large proteins that were newly created by the molecules in the mixture.

Based on what we know about translation, we know that proteins were produced in the **positive control** test tube, which contained all of the components, and the **second column** test tube, which contained everything except chromosomes. Chromosomes were not needed for translation because the mRNA was already in the mixture.

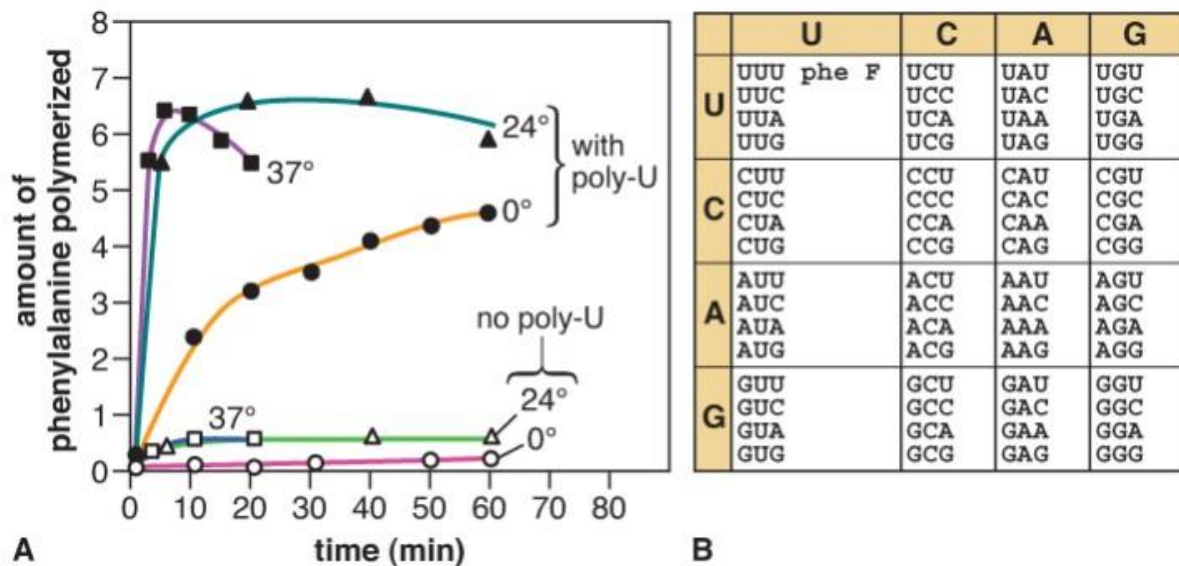
For additional information on translation, check out this video!

Translation:

https://www.youtube.com/watch?v=l3ayESylld0&list=PLYjFOc4FIyikoVfco6zaxMx5_CDDHj2m&index=75

HIGHLIGHT #1: Translation of RNA to Protein

In order to understand how RNA codons were translated to protein, scientists wrote down a table of all **64 RNA nucleotide triplets (codons)** and began to experimentally determine which amino acids were encoded by which codons. Mathematics dictated that some of the 20 amino acids would be encoded by more than one codon, since there are over three times as many codons as amino acids. To begin deciphering the genetic code, they synthesized a polymer of uracil-containing nucleotides, UUUUUUU..., which was added to an in-vitro translation mixture. **They conducted the experiment at three different temperatures because the in-vivo rate of translation of naturally produced mRNA was known for these temperatures.** They also conducted negative control experiments that omitted mRNA polymers of uracil, also called poly-U. The results are below.



As you can see, based on their results, the investigators determined that phenylalanine had been polymerized into an artificial protein, which cracked the genetic code for the first codon: **UUU = phe or F.**

Notice that without the poly-U, almost no phenylalanine was made, but in the treatments with poly-U, the production was significantly higher. This makes sense, because it shows us that the UUU codon is needed for Phenylalanine to be polymerized.

HIGHLIGHT #2: The Role of Temperature

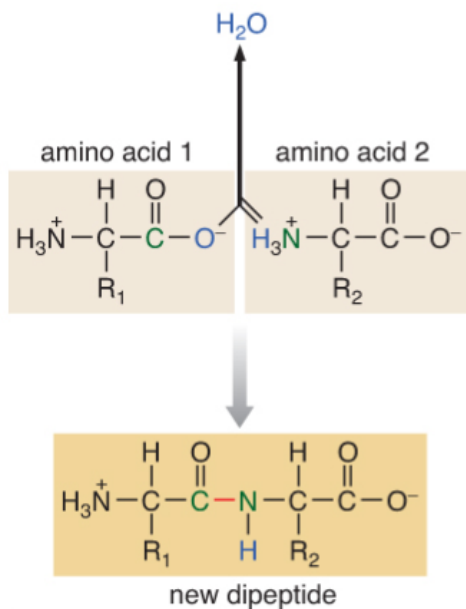
In the previous experiment, we can see that at 0 degrees, there is some production of phenylalanine, which increases at 24 degrees, but polymerization of phenylalanine **completely stops** at 37 degrees after a certain amount of time.

Why did this happen?

Biological processes are often **temperature-dependent**. Many processes increase in speed when temperature increases as a result of increased enzyme activity and reaction rates. However, if the temperature continues to increase, after a certain point, the temperature is too high for molecules to maintain their structure. This can cause proteins to denature, or unfold. Of the three temperatures which were included in the above experiment, 24 degrees was closest to the **optimum temperature** for this reaction.

HIGHLIGHT #3: Peptide Bonds

Amino acids are connected to each other via **peptide bonds** to make a **polypeptide**. **Peptide bonds** are covalent bonds and are catalyzed by specific enzymes. The ribosome helps to position these enzymes, the mRNA, and the amino acids correctly so that this process can occur.



As you can see, when two amino acids are brought together, a water molecule is released when the bond is created. This is called a **dehydration** or **condensation** reaction, and it requires energy. When a peptide bond is created between two amino acids, a dipeptide is created. Each time an amino acid is added to the growing peptide chain, a new peptide bond is formed.

CHECK YOUR LEARNING

(Answers Below)

- 1) Where can each type of RNA be found in the cell?
- 2) Rank the three types of RNA from most abundant to least abundant.
- 3) Identify the amino acid sequence that would be generated from the below three codons.

AUGAUCUCG

(Hint: You will have to use a codon chart! These can be found anywhere online. There should be three amino acids total in the sequence).

- 4) For the first experiment, if the experimenters had decided to include a negative control, how would they have set up this test tube? What would this extra control group have shown them?

THINGS YOU MAY STRUGGLE WITH

- When a peptide bond is created, the water molecule that is released was not originally in the form of H₂O in the amino acids; instead, the H₂O molecule is a combination of **a hydrogen and oxygen which are released from one amino acid, and another hydrogen which is released from the other amino acid**. Two hydrogens plus one oxygen gives us H₂O, or water.
- When reading the first experiment with the seven test tubes, remember that **chromosomes are still needed in the cell for mRNA and protein production**. Translation was able to occur without chromosomes because the reaction mixture included mRNA which had ALREADY been transcribed from DNA; in-vivo, chromosomes are still essential for transcription, which needs to happen in order for translation to occur.
- In the poly U experiment, investigators included **two variables** in their experimental design. One variable was **the presence (or absence) of the poly U tail**, and the other variable was the **temperature** at which polymerization occurred.

ANSWERS

- 1) rRNA can be found in the ribosomes in the cytoplasm, mRNA can be found in the nucleus OR in the cytosol, and tRNA can be found in the cytosol of the cell.
- 2) rRNA, tRNA, mRNA
- 3) Methionine-Isoleucine-Serine
- 4) They could have set up an additional test tube that contained only a buffer and none of the other molecules. This would have confirmed that the proteins which showed up on the gel were newly polymerized by the ingredients in the test tube and were not initially in the mixture.

Thanks for checking out these weekly resources! Don't forget to check out our website for group tutoring times, video tutorials and lots of other resources: www.baylor.edu/tutoring! Answers to Check your Learning questions are below!