Translation: Nucleic Acid to Polypeptide

7.3

Transcription uses the message coded in a DNA strand as a template for the synthesis of a complementary single-stranded mRNA molecule. The next step in the process of manufacturing a protein is translating the information coded in mRNA to a protein. In translation, the encoded message is read, codon by codon, by a ribosome and, with the presence of transfer RNA molecules, the ribosome assembles one amino acid at a time into a polypeptide chain.

tRNA

The ribosome alone cannot synthesize the polypeptide chain. Transfer RNA molecules (tRNAs) are small RNAs, about 70 to 90 nucleotides long. In comparison, mRNAs are typically hundreds of nucleotides long. tRNAs have a highly distinctive structure that serves their role in translation (**Figure 1**). All tRNAs have regions that base pair with themselves, winding into four double-helical segments to form a cloverleaf pattern. At the tip of one of the double-helical segments is an **anticodon**: a 3-nucleotide segment that pairs with a codon in an mRNA. At the other end of the cloverleaf is a region that carries the amino acid that corresponds to the anticodon. For example, a tRNA that is linked to serine (Ser) pairs with the codon 5′-AGU-3′ in mRNA. The anticodon of the tRNA that pairs with this codon is 3′-UCA-5′.

anticodon the complementary sequence of base pairs on a tRNA that corresponds to a codon on an mRNA

Recall that 61 of the 64 codons of the genetic code specify an amino acid. However, this does not mean that we need 61 different tRNAs to read the different codons. Francis Crick's wobble hypothesis proposed that the complete set of 61 codons can be read by fewer than 61 distinct tRNAs because of the particular pairing properties of the bases in the anticodons. That is, the pairing of the anticodon with the first two nucleotides of the codon is always precise, but most anticodons have flexibility in pairing with the third nucleotide of the codon. For example, UAU and UAC both code for tyrosine. If the tRNA's anticodon is AUA, it can still bind to the codon UAC, despite its complementarity being UAU. Either way, tyrosine is added on (**Figure 2**).

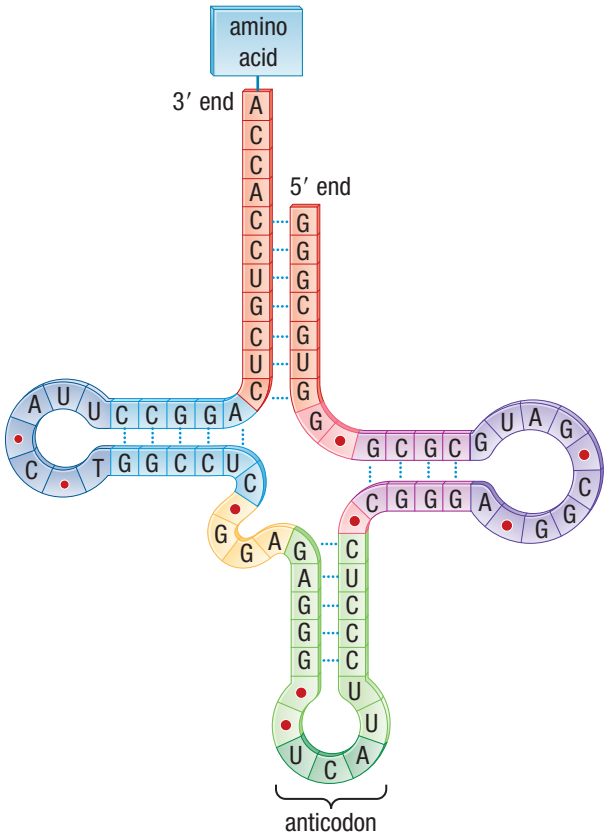

Figure 1 The structure of tRNA: The red dots show sites where bases are chemically modified into other forms.

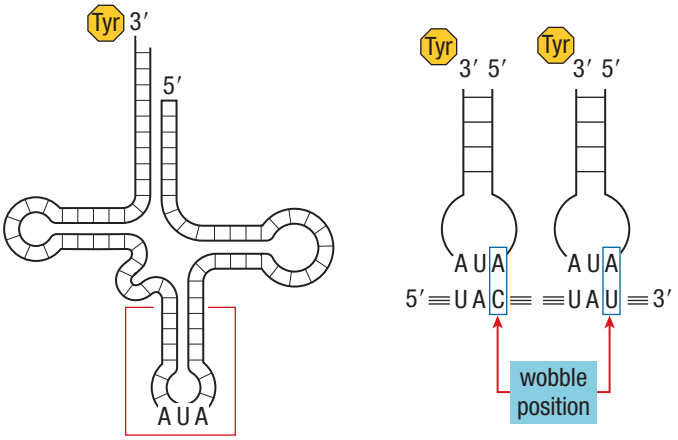

Figure 2 The wobble hypothesis: Even though the third nucleotide in the mRNA differs (UAU and UAC), the same tRNA with the anticodon AUA will deliver tyrosine to the growing polypeptide chain. Therefore, fewer types of tRNA are required to deliver the 20 amino acids, even though 61 codons exist in the genetic code.

aminoacylation the process by which a tRNA molecule is bound to its corresponding amino acid

aminoacyl–tRNA a molecule of transfer RNA bound to its associated amino acid

The process of adding an amino acid to a tRNA is called **aminoacylation** (literally, the addition of an amino acid) or "charging" the tRNA. The finished product, a tRNA linked to its correct amino acid, is called an **aminoacyl–tRNA**. Aminoacylation is catalyzed by 20 different aminoacyl–tRNA synthetase enzymes, one for each of the 20 amino acids. The energy in the aminoacyl–tRNA eventually drives the formation of the peptide bond that links the amino acids during translation.

With the tRNAs attached to their corresponding amino acids, our attention moves to the ribosome, where the amino acids are removed from tRNAs and linked together into polypeptide chains.

Ribosomes

Ribosomes carry out protein synthesis by translating mRNA into chains of amino acids. Like automated machines that use a series of steps to form complicated metal or plastic parts, ribosomes use an information tape—an mRNA molecule—to accomplish a task. For ribosomes, the task is joining amino acids in ordered sequences to make polypeptide chains.

A ribosome is made up of two different-sized parts, called the large and small ribosomal subunits (**Figure 3**). Each subunit is made up of a combination of ribosomal RNA (rRNA) and ribosomal proteins. To fulfill its role in translation, the ribosome has special binding sites that actively bring together mRNA with aminoacyl–tRNAs (Figure 3). One such site is where the mRNA threads through the ribosome. The A (aminoacyl) site is where the incoming aminoacyl–tRNA, carrying the next amino acid to be added to the polypeptide chain, binds to the mRNA. The P (peptidyl) site is where the tRNA, carrying the growing polypeptide chain, is bound. The E (exit) site is where an exiting tRNA leaves the ribosome.

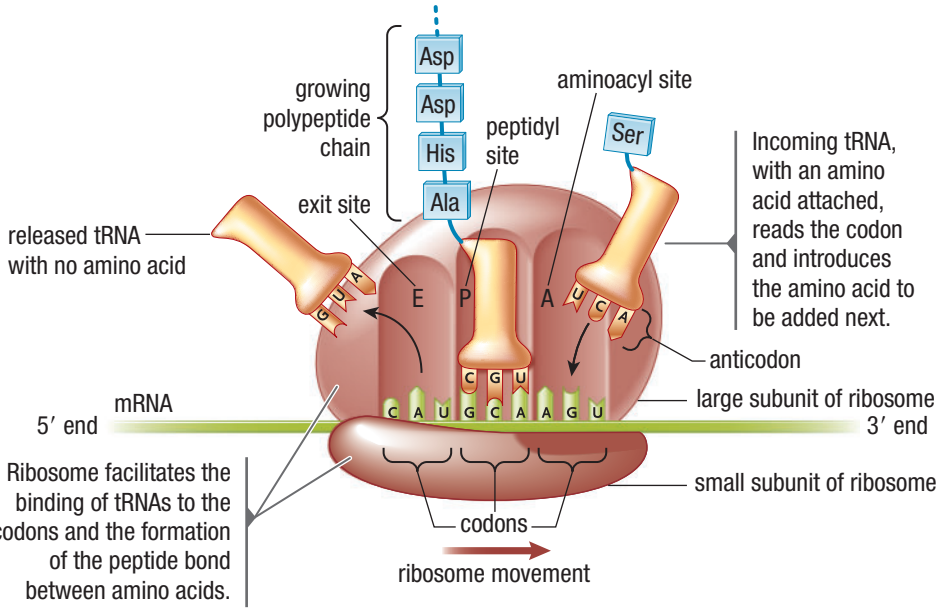

Figure 3 A ribosome assembles amino acids into a polypeptide chain. A tRNA molecule, with an amino acid bound to it, enters the ribosome on the right (A site). The anticodon on the tRNA pairs with the codon in the mRNA. Its amino acid will then be added to the growing polypeptide, which is currently attached to the tRNA in the middle of the ribosome (P site).

The Process of Translation

There are three major stages of translation: initiation, elongation, and termination. The binding of the ribosome subunits to the mRNA molecule and the recognition of the start codon initiates translation. The start codon AUG corresponds to the amino acid methionine. The polypeptide chain is elongated as the codons (sets of three

nucleotides) are read. tRNA molecules deliver the appropriate amino acids to the ribosome, where they are added to the end of the growing chain. The termination process recognizes where to stop adding amino acids to the growing polypeptide. A stop codon terminates translation. Multiple ribosomes along the same mRNA can simultaneously translate the mRNA and make multiple copies of the protein.

Initiating Translation

The first stage in assembling a protein, using mRNA as a template, is called initiation. Translation is initiated when the large and small ribosomal subunits associate with an mRNA molecule, and the first aminoacyl–tRNA of the new protein chain becomes bound to the AUG start codon. The aminoacyl–tRNA that is used for initiation is a specialized initiator tRNA, which has an anticodon to the methionine-specifying AUG start codon.

In Step 1 of the initiation process, the initiator methionine–tRNA (met–tRNA) forms a complex with the small ribosomal subunit (**Figure 4**). The complex binds to the mRNA at the 5′ cap and then moves along the mRNA (a process called scanning) until it reaches the first AUG codon (Step 2). This is the start codon, and it is recognized by the anticodon of the Met–tRNA. The large ribosomal subunit then binds to complete the ribosome (Step 3). At the end of initiation, the initiator Met–tRNA is in the P site.

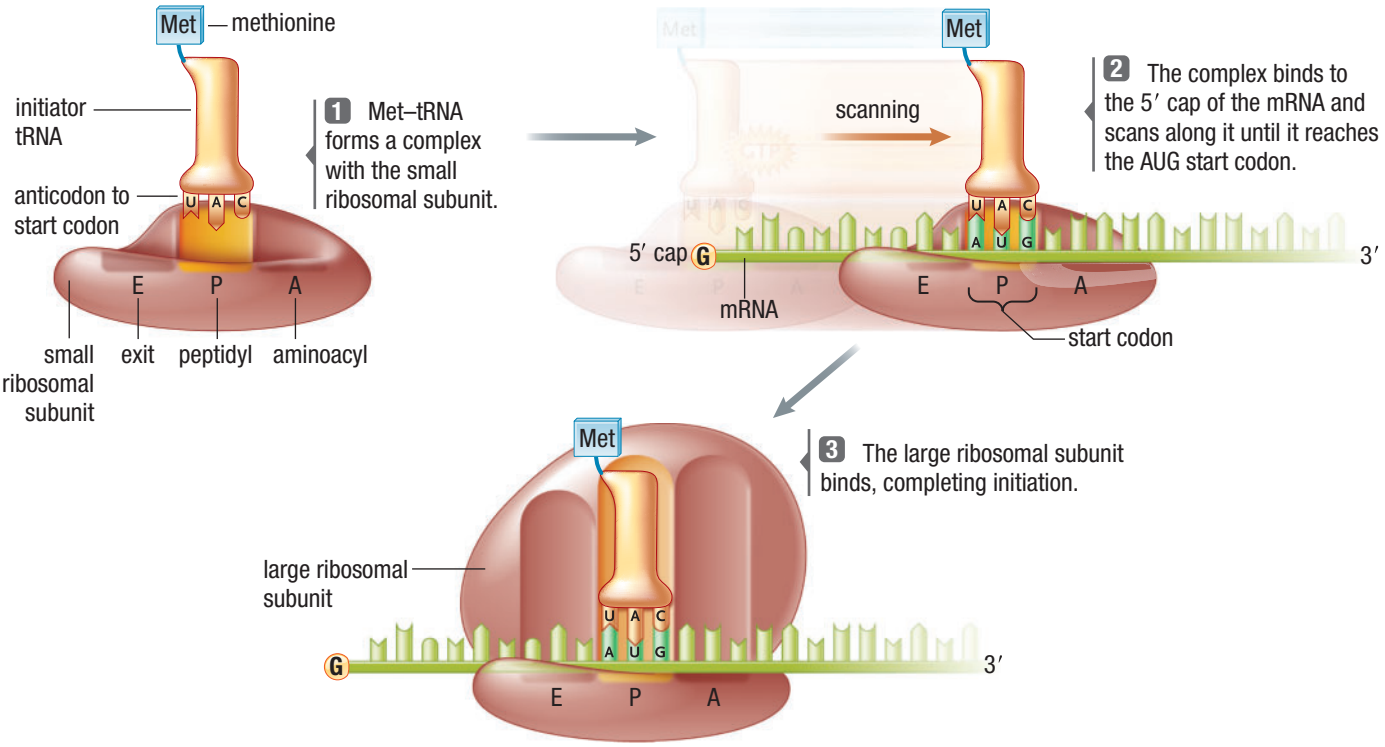

Figure 4 The steps in the initiation stage of translation in eukaryotes

After the initiator tRNA pairs with the AUG initiator codon, the subsequent stages of translation simply read the nucleotide bases, three at a time, on the mRNA. Since each codon consists of three bases, a sequence could potentially be read in three different ways, depending on where the ribosome begins. A correct initiator tRNA–AUG pairing establishes the correct **reading frame**: the series of codons for the polypeptide that is encoded by the mRNA.

reading frame a particular system for separating a base pair sequence into readable codons

Elongating the Polypeptide Chain

The central reactions of translation take place in the four steps of the elongation stage, which adds single amino acids sequentially to a growing polypeptide chain. **Figure 5** (next page) shows how the A, P, and E sites operate through the elongation stage. Elongation begins when an initiator tRNA, with its attached methionine, is bound to the P site in Step 1. The A site is empty.

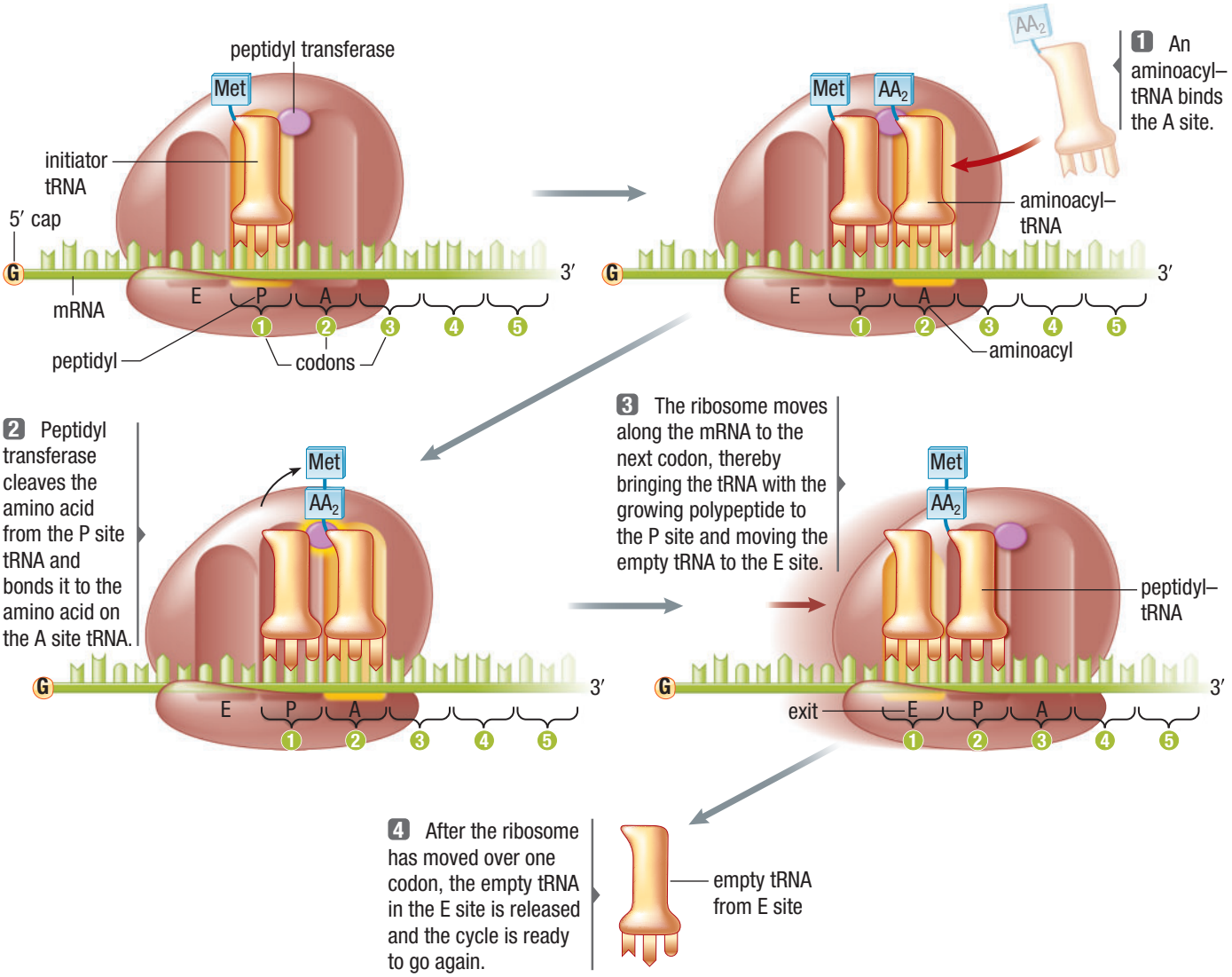

Figure 5 The steps in the elongation stage of translation

In Step 2, the second tRNA, with an appropriate anticodon and amino acid (AA_2), binds to the codon in the A site of the ribosome. A GTP rather than an ATP is hydrolyzed to provide free energy for this step. Next, the amino acid (Met) is cleaved from the tRNA in the P site and forms a peptide bond with the amino acid (AA_2) on the tRNA in the A site. This bond formation is catalyzed by peptidyl transferase, which is a ribosomal enzyme. At the end of Step 2, the new polypeptide chain is attached to the tRNA in the A site and an empty tRNA remains at the P site.

In Step 3, the ribosome moves along the mRNA to the next codon. The two tRNAs remain bound to their respective codons, so this step positions the newly formed peptidyl–tRNA in the P site and generates a vacant A site. An appropriate tRNA moves into the A site, and Steps 2 and 3 are repeated. After each repeat, the empty tRNA that was in the P site moves to the E site. In Step 4, the empty tRNA is released from the ribosome.

Termination of Protein Synthesis

Translation switches from the elongation to the termination stage when the A site of a ribosome arrives at one of the stop codons (UAA, UAG, or UGA) on the mRNA. When a stop codon appears at the A site, a protein release factor binds at this site instead of an aminoacyl–tRNA. In response, the polypeptide chain is released from the tRNA at the P site as usual. However, because no amino acid is present at the A site, the freed polypeptide chain is detached from the ribosome. At the same time, the ribosomal subunits separate and detach from the mRNA. The empty tRNA and the release factor are also released.

Investigation 7.3.1

Protein Synthesis (p. 354)
In this investigation, you will use your knowledge of transcription, translation, and the genetic code to analyze the genes and traits of a fictitious animal.

Protein Synthesis in Eukaryotes and in Prokaryotes

In both prokaryotes and eukaryotes, multiple ribosomes can translate an mRNA molecule at the same time, thereby increasing the production of crucial proteins (**Figure 6**). The complex that is formed is called a **polysome** (**Figure 7**).

polysome a complex that is formed when multiple ribosomes attach to the same mRNA molecule in order to facilitate rapid translation

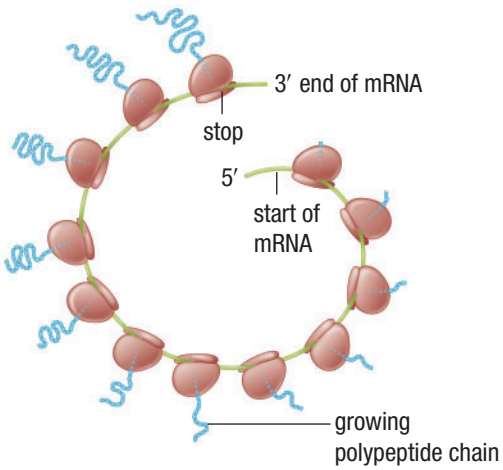

Figure 6 Polysomes consist of a series of ribosomes translating the same mRNA.

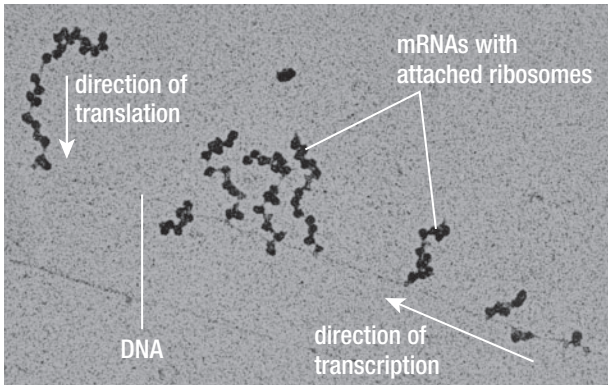

Figure 7 Simultaneous transcription and translation in progress in a prokaryotic cell. This electron microscope preparation was extracted from *E. coli* (magnified × 5 700 000).

In eukaryotes, polysomes only form outside the nucleus, in the cytosol. In prokaryotes, however, transcription and translation both occur in the cytosol. Thus the ribosomes have access to mRNA even as it is being synthesized. As a result, protein synthesis can occur at a much higher rate in prokaryotes than in eukaryotes. The tight coupling of transcription and translation also allows prokaryotes to rapidly synthesize proteins in response to changing environmental conditions. Other key differences are summarized in **Table 1**.

Table 1 A Comparison of Translation in Prokaryotes and Eukaryotes

Variable	Prokaryotes	Eukaryotes
location	• mRNA is translated by ribosomes in the cytosol as it is being transcribed from DNA	• mRNA can only be translated after exiting the nucleus to interact with ribosomes in the cytosol • some translation occurs in mitochondria and chloroplasts
initiation	• mRNA bases pair directly with a ribosomal binding site, just upstream of the start codon • mRNA 5′ cap is involved	• complex of Met–tRNA, with small ribosomal subunits, binds to an mRNA 5′ cap and scans until it encounters the start codon
elongation	• 15 to 20 elongation cycles per second	• 1 to 3 elongation cycles per second
termination	• stop codon appears and a release factor binds so that the polypeptide is released	
polysomes	• mRNA strand can be translated by multiple ribosomes simultaneously, even as it is being transcribed from DNA	• mRNA strand can be translated by multiple ribosomes simultaneously, but only in the cytosol

Polypeptide to Protein

The polypeptide that has been assembled by the ribosome is still not functional. The protein exists in an inactive state. Since the shape of a protein defines its function, the polypeptide chain must be folded into the correct conformation. Multiple processing reactions, carried out by specific enzymes, remove amino acids from the ends or interior of the chain and may add additional molecules, such as sugars, to the chain. These reactions activate the polypeptide, which then folds into its functional shape. In addition, many proteins are composed of two or more polypeptide chains. In these cases, the polypeptides produced from a number of separate translation events are processed and then assembled together to form a single functioning protein. This process of protein processing, from an inactive to an active state, is one of the many mechanisms that a cell uses to control the expression of its genes. You will learn more about gene expression in the next section.

Mini **Investigation**

Computer Simulation of Protein Synthesis

Skills: Performing, Observing, Analyzing, Evaluating, Communicating

SKILLS HANDBOOK A2.1

In this investigation, you will transcribe DNA and translate the information into a polypeptide.

1. Connect to the Internet, and explore the protein synthesis simulation suggested by your teacher.
2. Follow the simulation instructions to model the steps of transcription and translation that take place during protein synthesis.
3. Carefully observe the actions of the many molecules involved and note the roles of various enzymes.

A. Compare the steps modelled in the computer simulation with those you have learned in Sections 7.2 and 7.3. K/U

B. List the codons that signal a ribosome to stop building a polypeptide chain. T/I

C. How would you change this simulation to make transcription and translation more understandable? T/I

D. Computer simulations are often used to help visualize a cellular process. Based on your experience with the simulation of protein synthesis, comment on the usefulness of this approach. Did the animation help you understand the process of protein synthesis? T/I

WEB LINK

7.3 Review

Summary

- Translation is the assembly of amino acids into polypeptides by a ribosome, using the information encoded in mRNA. This process has three stages: initiation, elongation, and termination. It is dependent on tRNA molecules.
- tRNA molecules are small RNA molecules. Each tRNA is associated with a specific amino acid, as dictated by its respective anticodon.
- An anticodon is a triplet of complementary bases that is able to bind to mRNA codons that code for the tRNA's specific amino acid.
- Initiation of translation is triggered by the large and small ribosome subunits binding to the 5′ cap of an mRNA molecule. The start codon is AUG, which corresponds to the amino acid methionine. The tRNA whose anticodon is UAC delivers methionine to the ribosome.
- Elongation involves building the polypeptide chain. Incoming tRNA molecules deliver the next appropriate amino acid to the ribosome, as dictated by the reading frame of three bases.
- Termination occurs when a stop codon is read by the ribosome. The stop codons are UAG, UGA, and UAA.
- More than one ribosome can translate an mRNA molecule at a time. The resulting structure is called a polysome.

Questions

1. What are the key steps in the initiation of translation in eukaryotes and prokaryotes? K/U
2. What is the role of tRNA in translation? K/U
3. Why is there not a specific tRNA molecule for each possible codon? K/U
4. List the possible anticodons for phenylalanine, alanine, and tyrosine. K/U
5. The wobble hypothesis states that there is increased flexibility in base pairing at the third nucleotide of some codons. Why does this not lead to frequent mistakes in the assembly of proteins? T/I
6. Explain what occurs at the A, P, and E sites during the translation of mRNA into a polypeptide. K/U
7. What are some examples of processes that might demand high rates of protein synthesis in humans? T/I
8. Using a diagram, summarize the process of translation. Label the areas of initiation, elongation, and termination. Present your diagram to a classmate, in a format of your choice. C
9. The set of all proteins expressed by a genome over the lifetime of a cell is called the proteome. The proteome is constantly changing as proteins interact with other proteins and chemical signals inside and outside the cell. Scientists are turning to proteomics—the study of the structure, activities, and functions of proteins—to understand the molecular basis of health and disease. Using the Internet or other sources, research the Proteome Project. Find out about its mission, the people and organizations involved, the technologies being used, and current findings. Present your findings in a format of your choice. CAREER LINK K/U C
10. Many proteins are assembled from more than one kind of polypeptide. A hemoglobin protein, for example, consists of two alpha and two beta polypeptides. In this case, two genes are needed to code for a single protein. Do online research to find examples of other proteins that are coded for by multiple genes. Summarize the information you find. A

WEB LINK