

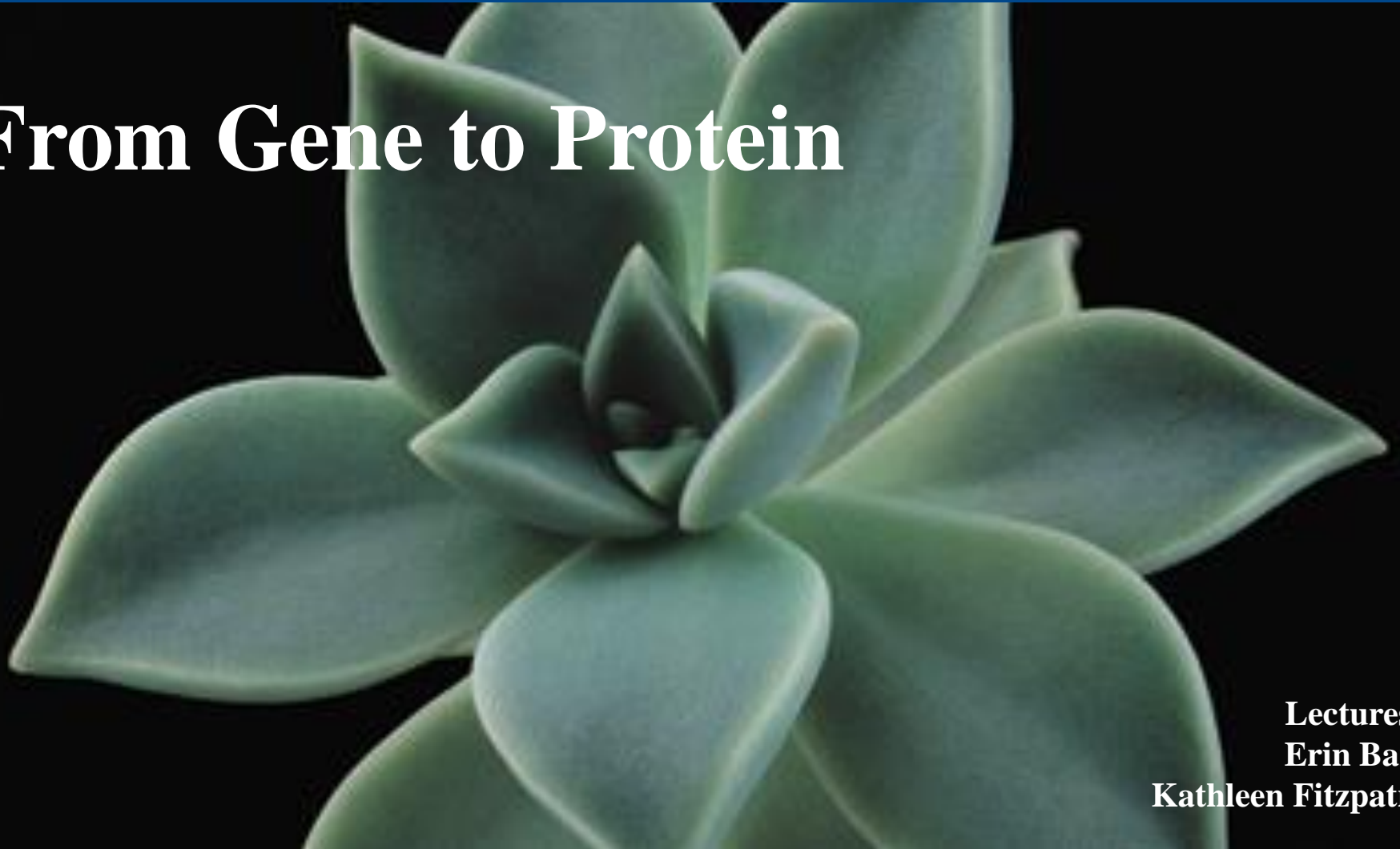
LECTURE PRESENTATIONS

For CAMPBELL BIOLOGY, NINTH EDITION

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Chapter 17

From Gene to Protein



Lectures by
Erin Barley
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Overview: The Flow of Genetic Information

- The information content of DNA is in the form of specific sequences of nucleotides
- The DNA inherited by an organism leads to specific traits by dictating the synthesis of proteins
- Proteins are the links between genotype and phenotype
- **Gene expression**, the process by which DNA directs protein synthesis, includes two stages: transcription and translation

Figure 17.1



Concept 17.1: Genes specify proteins via transcription and translation

- How was the fundamental relationship between genes and proteins discovered?

Evidence from the Study of Metabolic Defects

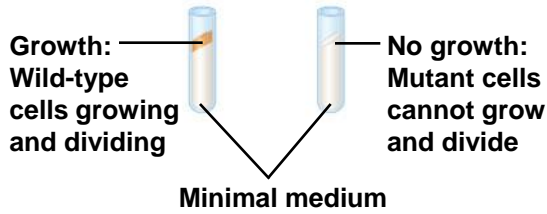
- In 1902, British physician Archibald Garrod first suggested that genes dictate phenotypes through enzymes that catalyze specific chemical reactions
- He thought symptoms of an inherited disease reflect an inability to synthesize a certain enzyme
- Linking genes to enzymes required understanding that cells synthesize and degrade molecules in a series of steps, a metabolic pathway

Nutritional Mutants in Neurospora: **Scientific Inquiry**

- George Beadle and Edward Tatum exposed bread mold to X-rays, creating mutants that were unable to survive on minimal media
- Using crosses, they and their coworkers identified three classes of arginine-deficient mutants, each lacking a different enzyme necessary for synthesizing arginine
- They developed a one gene–one enzyme hypothesis, which states that each gene dictates production of a specific enzyme

Figure 17.2

EXPERIMENT



RESULTS

		Classes of <i>Neurospora crassa</i>			
		Wild type	Class I mutants	Class II mutants	Class III mutants
Condition	Minimal medium (MM) (control)				
	MM + ornithine				
	MM + citrulline				
	MM + arginine (control)				
	Summary of results	Can grow with or without any supplements	Can grow on ornithine, citrulline, or arginine	Can grow only on citrulline or arginine	Require arginine to grow

CONCLUSION

Gene (codes for enzyme)	Wild type	Class I mutants (mutation in gene A)	Class II mutants (mutation in gene B)	Class III mutants (mutation in gene C)
Gene A → Enzyme A	Precursor Enzyme A	Precursor Enzyme A	Precursor Enzyme A	Precursor Enzyme A
Gene B → Enzyme B	Ornithine Enzyme B	Ornithine Enzyme B	Ornithine Enzyme B	Ornithine Enzyme B
Gene C → Enzyme C	Citrulline Enzyme C	Citrulline Enzyme C	Citrulline Enzyme C	Citrulline Enzyme C
	Arginine	Arginine	Arginine	Arginine

The Products of Gene Expression: A Developing Story

- Some proteins aren't enzymes, so researchers later revised the hypothesis: one gene—one protein
- Many proteins are composed of several polypeptides, each of which has its own gene
- Therefore, Beadle and Tatum's hypothesis is now restated as the one gene—one polypeptide hypothesis
- Note that it is common to refer to gene products as proteins rather than polypeptides

Basic Principles of Transcription and Translation

- RNA is the bridge between genes and the proteins for which they code
- **Transcription** is the synthesis of RNA using information in DNA
- Transcription produces **messenger RNA (mRNA)**
- **Translation** is the synthesis of a polypeptide, using information in the mRNA
- **Ribosomes** are the sites of translation

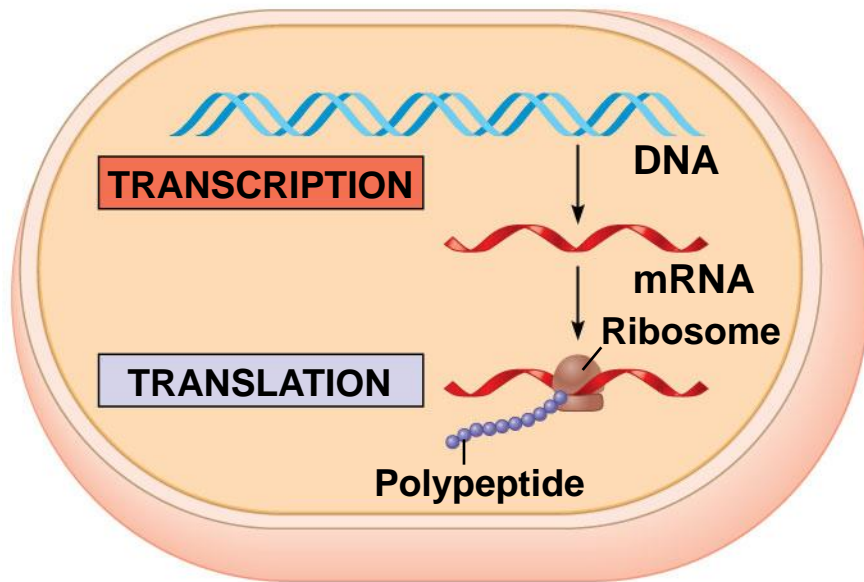
- In prokaryotes, translation of mRNA can begin before transcription has finished
- In a eukaryotic cell, the nuclear envelope separates transcription from translation
- Eukaryotic RNA transcripts are modified through RNA processing to yield the finished mRNA

- A **primary transcript** is the initial RNA transcript from any gene prior to processing
- The *central dogma* is the concept that cells are governed by a cellular chain of command:
DNA → RNA → protein

Figure 17.UN01

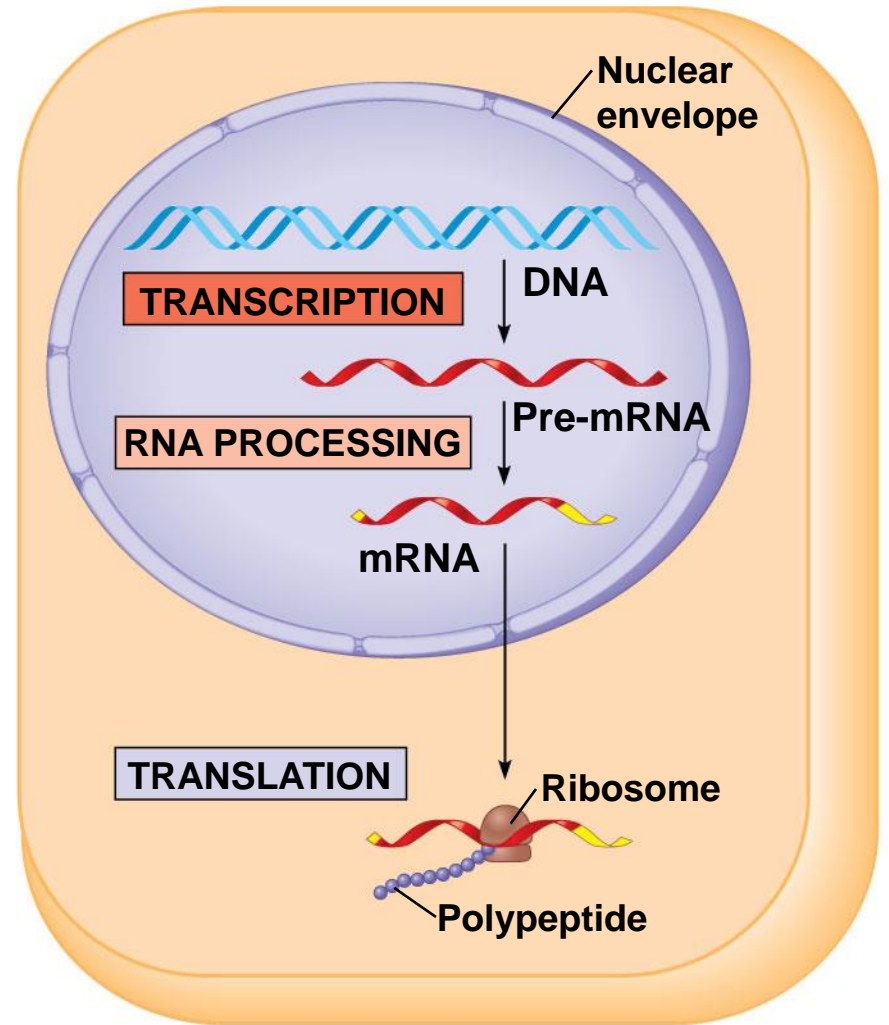


Figure 17.3



(a) Bacterial cell

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(b) Eukaryotic cell

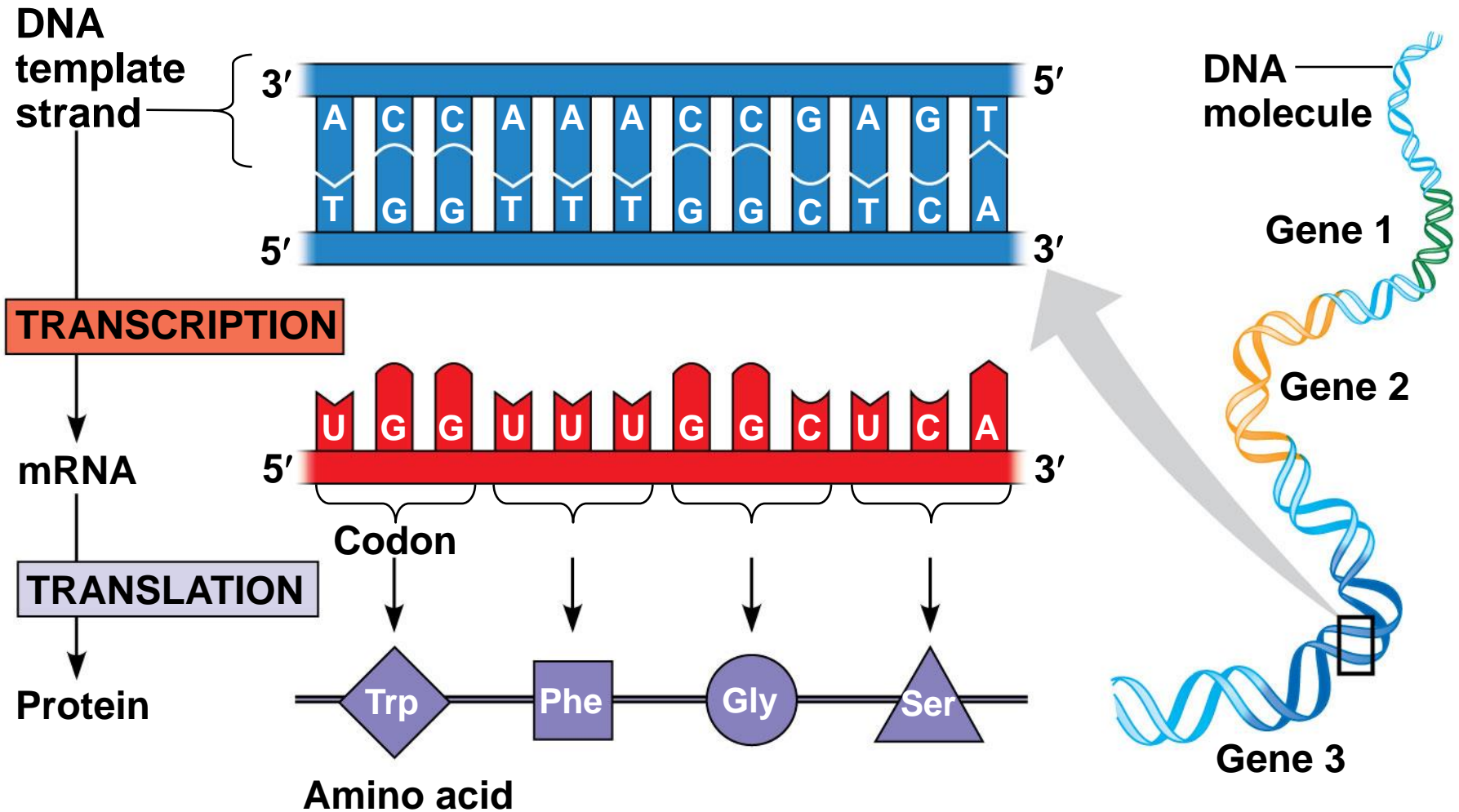
The Genetic Code

- How are the instructions for assembling amino acids into proteins encoded into DNA?
- There are 20 amino acids, but there are only four nucleotide bases in DNA
- How many nucleotides correspond to an amino acid?

Codons: Triplets of Nucleotides

- The flow of information from gene to protein is based on a **triplet code**: a series of nonoverlapping, three-nucleotide words
- The words of a gene are transcribed into complementary nonoverlapping three-nucleotide words of mRNA
- These words are then translated into a chain of amino acids, forming a polypeptide

Figure 17.4



- During transcription, one of the two DNA strands, called the **template strand**, provides a template for ordering the sequence of complementary nucleotides in an RNA transcript
- The template strand is always the same strand for a given gene
- During translation, the mRNA base triplets, called **codons**, are read in the 5' to 3' direction

- Codons along an mRNA molecule are read by translation machinery in the 5' to 3' direction
- Each codon specifies the amino acid (one of 20) to be placed at the corresponding position along a polypeptide

Cracking the Code

- All 64 codons were deciphered by the mid-1960s
- Of the 64 triplets, 61 code for amino acids; 3 triplets are “stop” signals to end translation
- The genetic code is redundant (more than one codon may specify a particular amino acid) but not ambiguous; no codon specifies more than one amino acid
- Codons must be read in the correct **reading frame** (correct groupings) in order for the specified polypeptide to be produced

Figure 17.5

		Second mRNA base						
		U	C	A	G			
First mRNA base (5' end of codon)	U	UUU	UCU	UAU	UGU	U	Third mRNA base (3' end of codon)	
		UUC	UCC	UAC	UGC			C
		UUA	UCA	UAA Stop	UGA Stop			A
		UUG	UCG	UAG Stop	UGG Trp			G
	C	CUU	CCU	CAU	CGU	U	Third mRNA base (3' end of codon)	
		CUC	CCC	CAC	CGC			C
		CUA	CCA	CAA	CGA			A
		CUG	CCG	CAG	CGG			G
	A	AUU	ACU	AAU	AGU	U	Third mRNA base (3' end of codon)	
		AUC	ACC	AAC	AGC			C
		AUA	ACA	AAA	AGA			A
		AUG Met or start	ACG	AAG	AGG			G
	G	GUU	GCU	GAU	GGU	U	Third mRNA base (3' end of codon)	
		GUC	GCC	GAC	GGC			C
		GUA	GCA	GAA	GGA			A
		GUG	GCG	GAG	GGG			G

Evolution of the Genetic Code

- The genetic code is nearly universal, shared by the simplest bacteria to the most complex animals
- Genes can be transcribed and translated after being transplanted from one species to another

Figure 17.6



(a) Tobacco plant expressing a firefly gene

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(b) Pig expressing a jellyfish gene

Concept 17.2: Transcription is the DNA-directed synthesis of RNA: *a closer look*

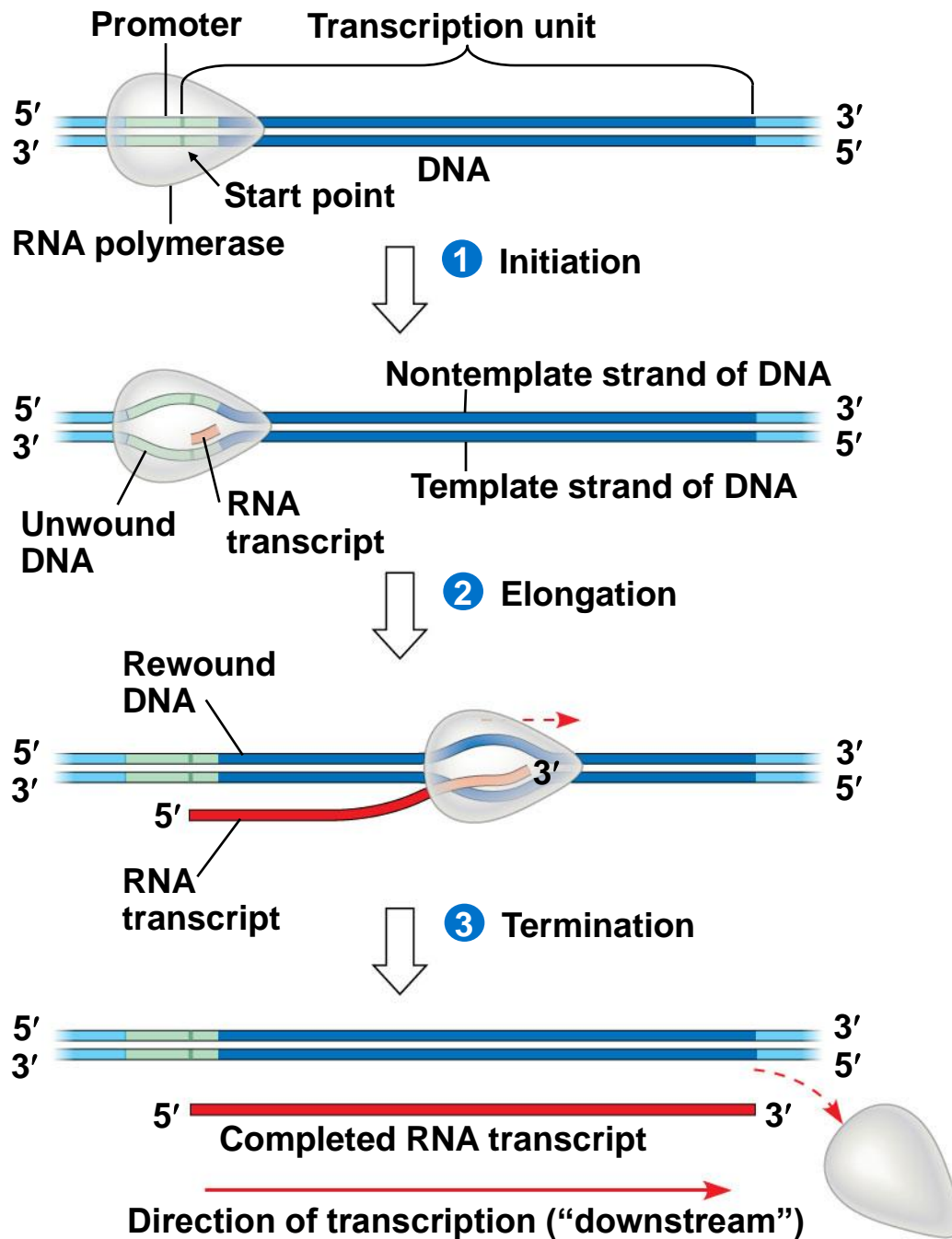
- Transcription is the first stage of gene expression

Molecular Components of Transcription

- RNA synthesis is catalyzed by **RNA polymerase**, which pries the DNA strands apart and hooks together the RNA nucleotides
- The RNA is complementary to the DNA template strand
- RNA synthesis follows the same base-pairing rules as DNA, except that uracil substitutes for thymine

- The DNA sequence where RNA polymerase attaches is called the **promoter**; in bacteria, the sequence signaling the end of transcription is called the **terminator**
- The stretch of DNA that is transcribed is called a **transcription unit**

Figure 17.7-4



Synthesis of an RNA Transcript

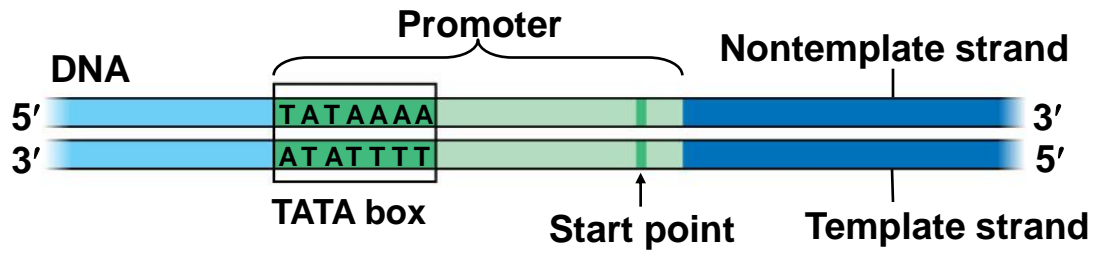
- The three stages of transcription
 - Initiation
 - Elongation
 - Termination

RNA Polymerase Binding and Initiation of Transcription

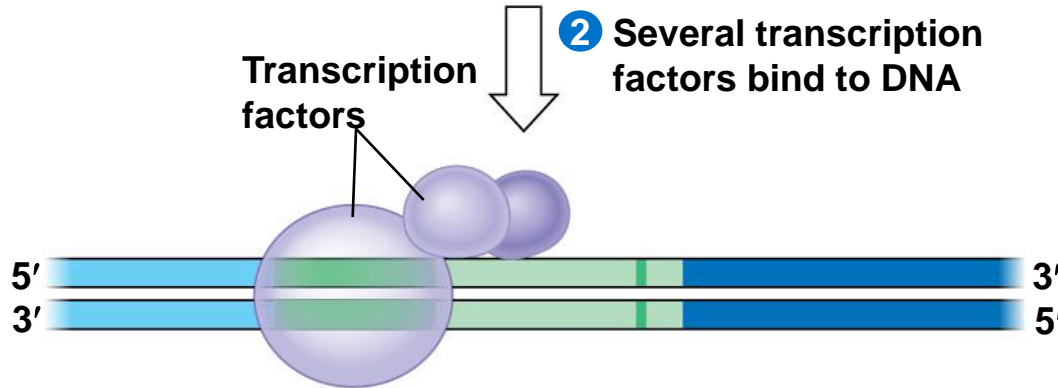
- Promoters signal the transcriptional **start point** and usually extend several dozen nucleotide pairs upstream of the start point
- **Transcription factors** mediate the binding of RNA polymerase and the initiation of transcription
- The completed assembly of transcription factors and RNA polymerase II bound to a promoter is called a **transcription initiation complex**
- A promoter called a **TATA box** is crucial in forming the initiation complex in eukaryotes

Figure 17.8

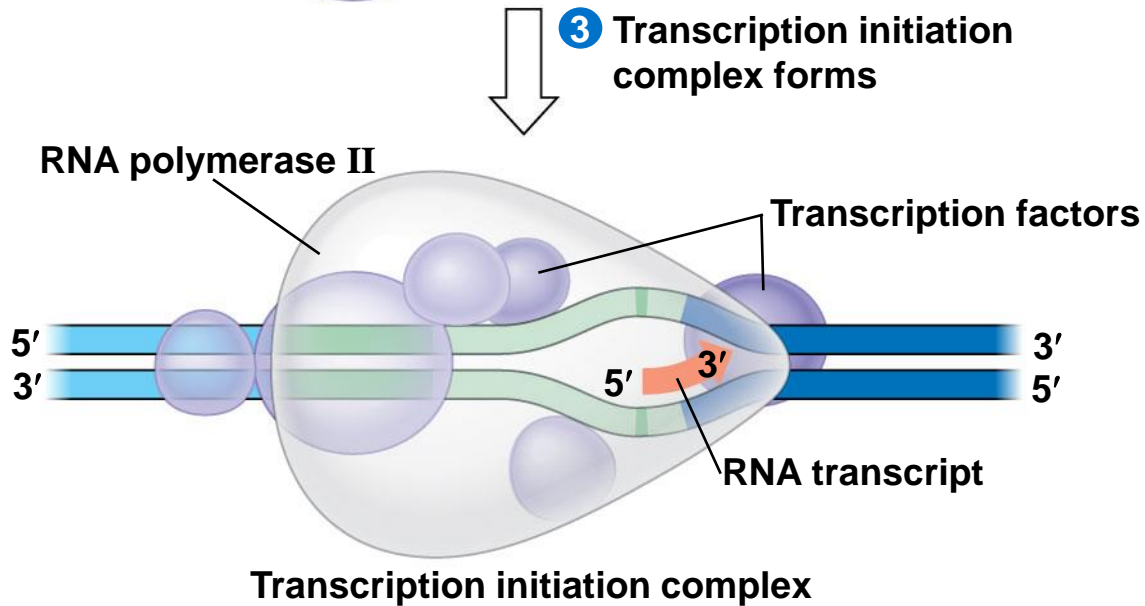
1 A eukaryotic promoter



2 Several transcription factors bind to DNA



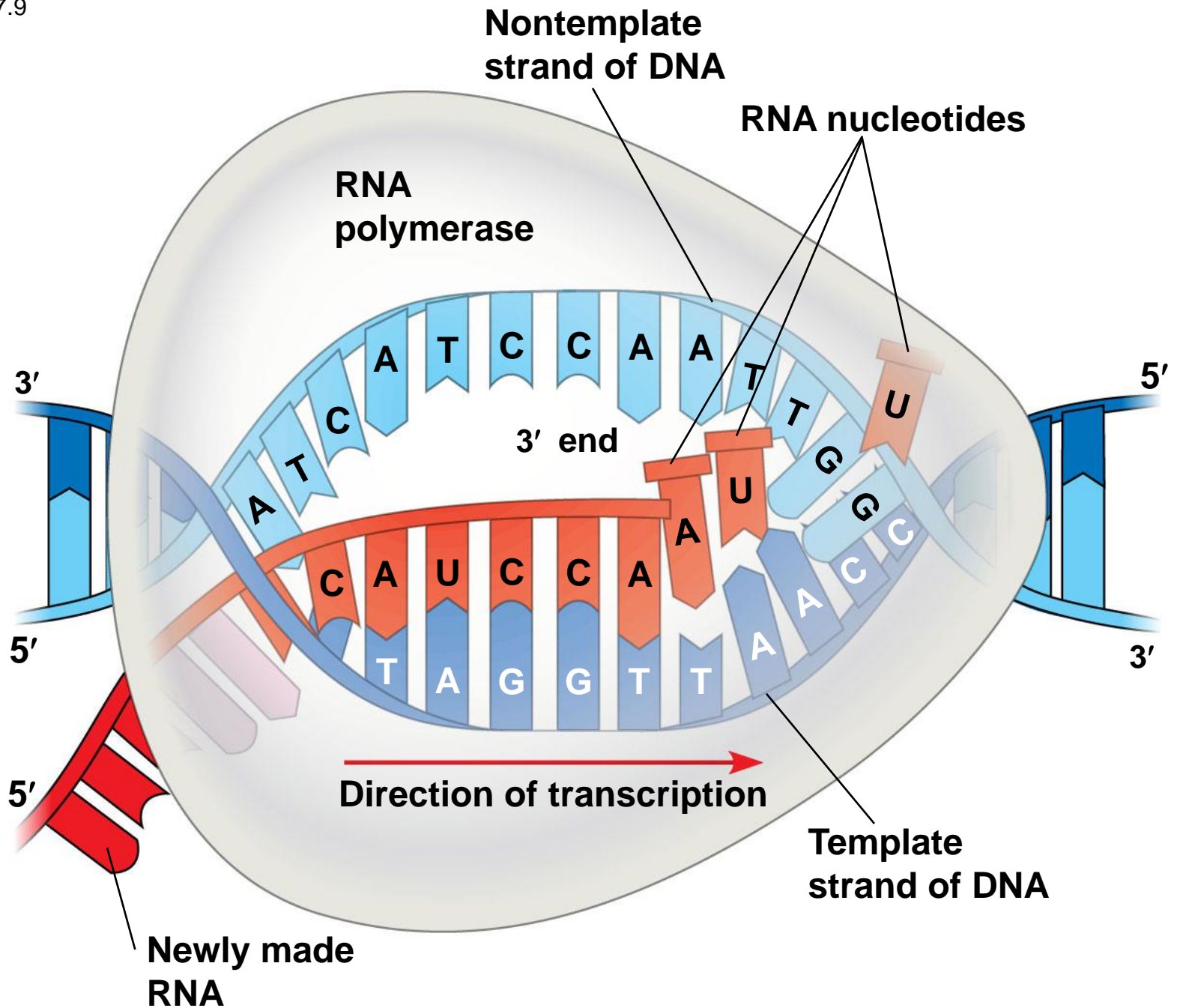
3 Transcription initiation complex forms



Elongation of the RNA Strand

- As RNA polymerase moves along the DNA, it untwists the double helix, 10 to 20 bases at a time
- Transcription progresses at a rate of 40 nucleotides per second in eukaryotes
- A gene can be transcribed simultaneously by several RNA polymerases
- Nucleotides are added to the 3' end of the growing RNA molecule

Figure 17.9



Termination of Transcription

- The mechanisms of termination are different in bacteria and eukaryotes
- In bacteria, the polymerase stops transcription at the end of the terminator and the mRNA can be translated without further modification
- In eukaryotes, RNA polymerase II transcribes the polyadenylation signal sequence; the RNA transcript is released 10–35 nucleotides past this polyadenylation sequence

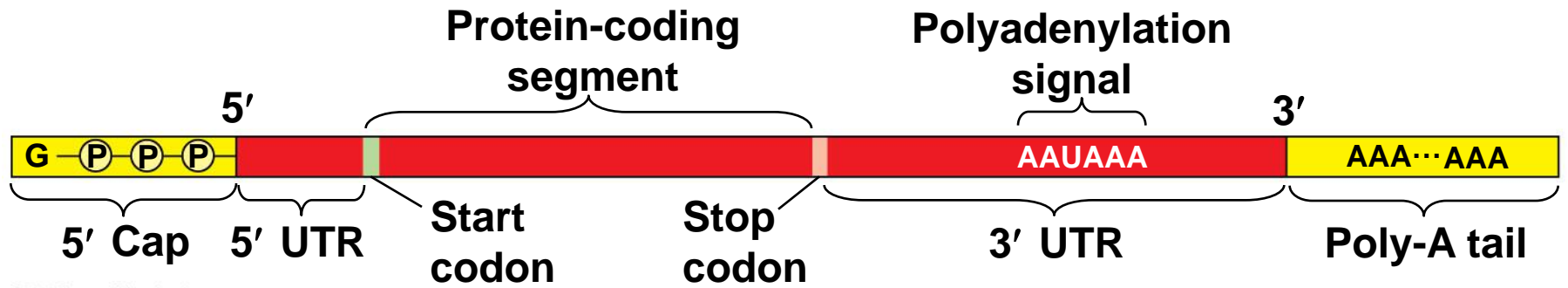
Concept 17.3: Eukaryotic cells modify RNA after transcription

- Enzymes in the eukaryotic nucleus modify pre-mRNA (**RNA processing**) before the genetic messages are dispatched to the cytoplasm
- During RNA processing, both ends of the primary transcript are usually altered
- Also, usually some interior parts of the molecule are cut out, and the other parts spliced together

Alteration of mRNA Ends

- Each end of a pre-mRNA molecule is modified in a particular way
 - The 5' end receives a modified nucleotide **5' cap**
 - The 3' end gets a **poly-A tail**
- These modifications share several functions
 - They seem to facilitate the export of mRNA to the cytoplasm
 - They protect mRNA from hydrolytic enzymes
 - They help ribosomes attach to the 5' end

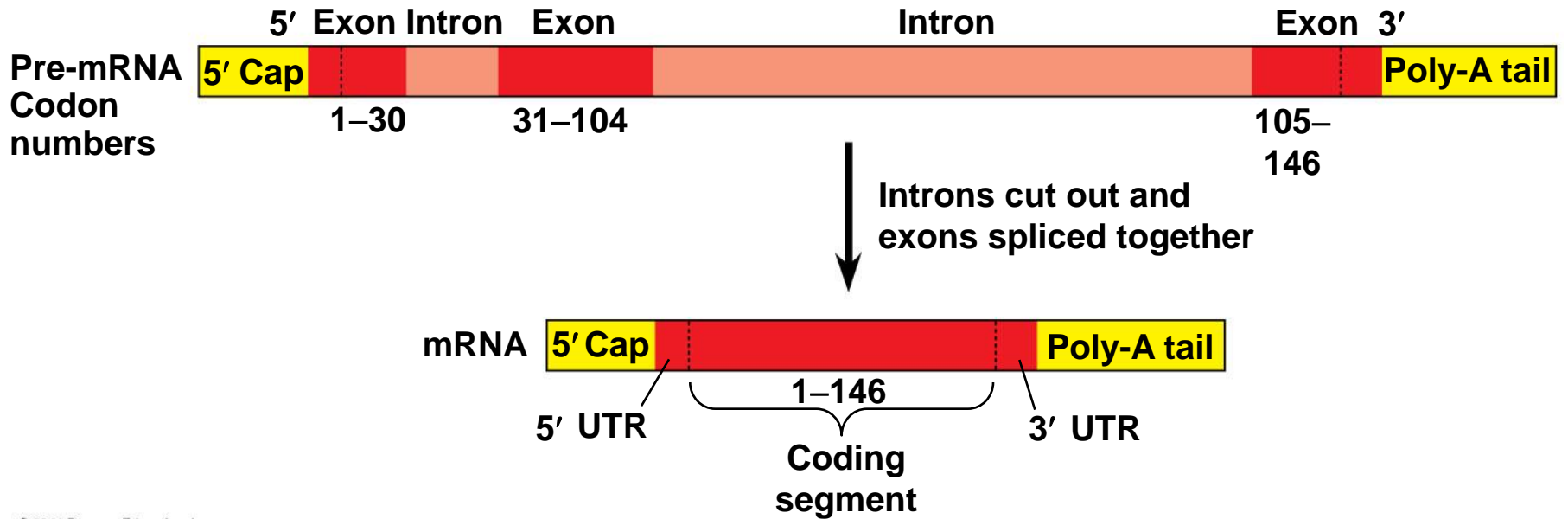
Figure 17.10



Split Genes and RNA Splicing

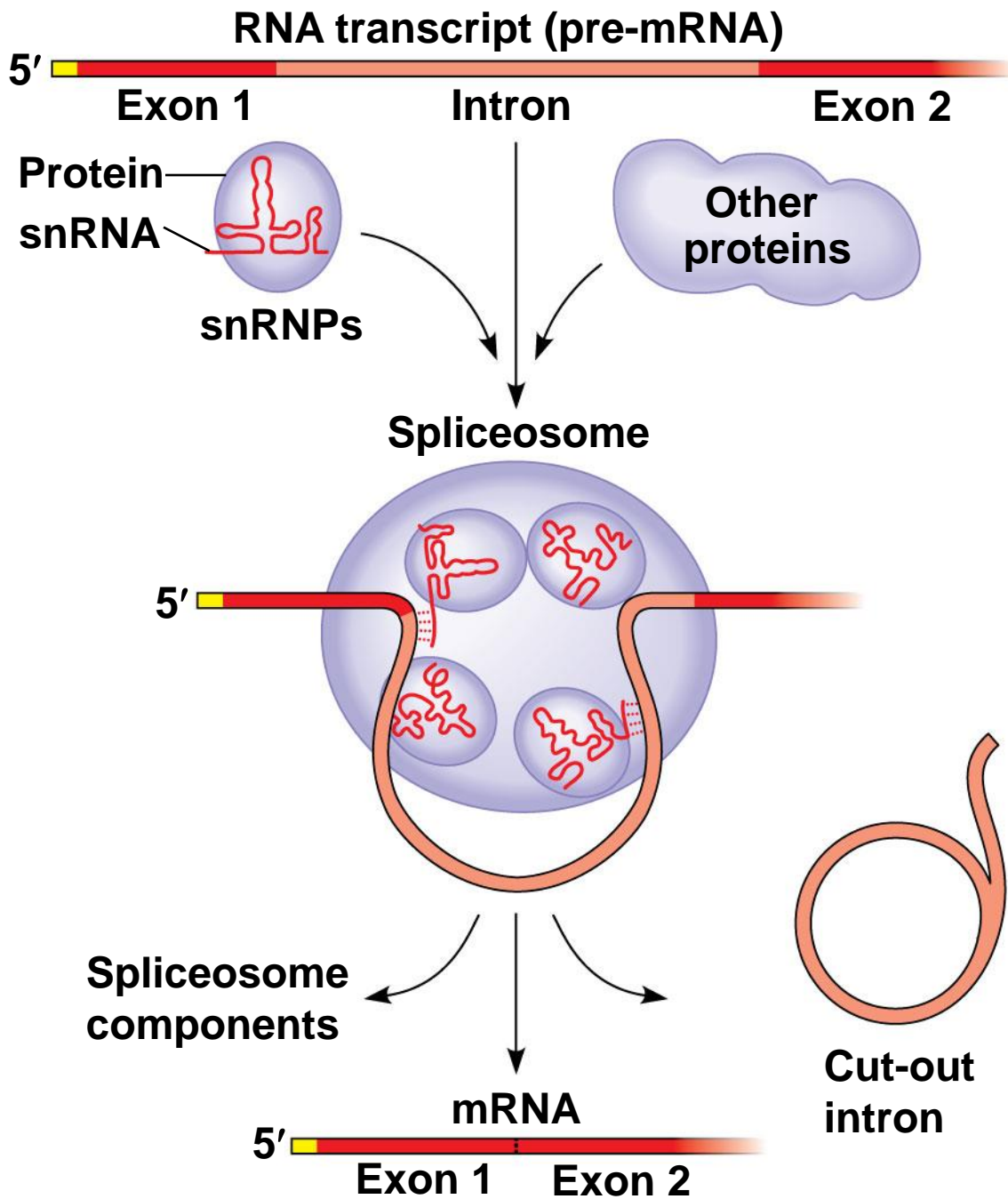
- Most eukaryotic genes and their RNA transcripts have long noncoding stretches of nucleotides that lie between coding regions
- These noncoding regions are called intervening sequences, or **introns**
- The other regions are called **exons** because they are eventually expressed, usually translated into amino acid sequences
- **RNA splicing** removes introns and joins exons, creating an mRNA molecule with a continuous coding sequence

Figure 17.11



- In some cases, RNA splicing is carried out by spliceosomes
- **Spliceosomes** consist of a variety of proteins and several small nuclear ribonucleoproteins (snRNPs) that recognize the splice sites

Figure 17.12-3



Ribozymes

- **Ribozymes** are catalytic RNA molecules that function as enzymes and can splice RNA
- The discovery of ribozymes rendered obsolete the belief that all biological catalysts were proteins

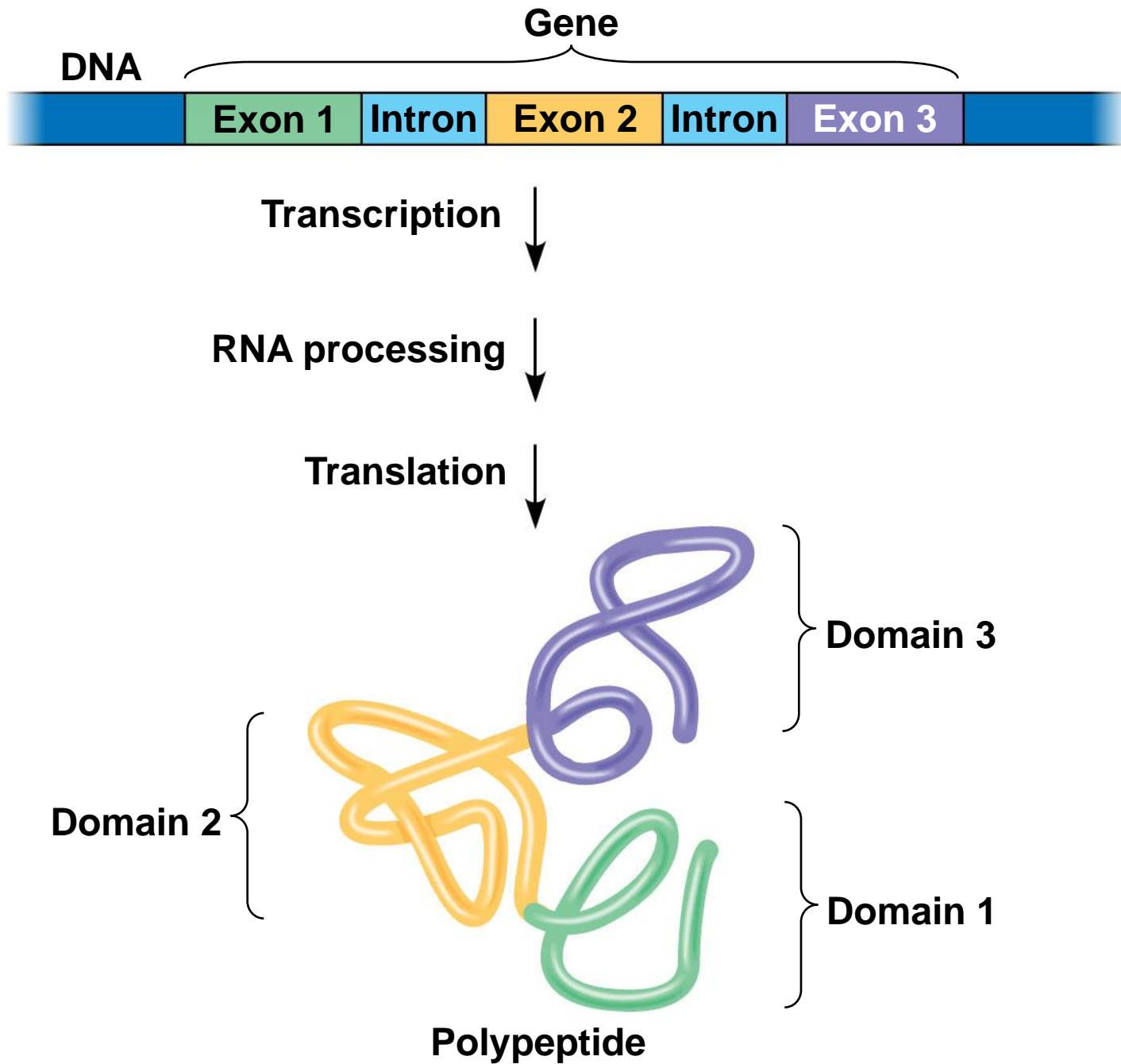
- Three properties of RNA enable it to function as an enzyme
 - It can form a three-dimensional structure because of its ability to base-pair with itself
 - Some bases in RNA contain functional groups that may participate in catalysis
 - RNA may hydrogen-bond with other nucleic acid molecules

The Functional and Evolutionary Importance of Introns

- Some introns contain sequences that may regulate gene expression
- Some genes can encode more than one kind of polypeptide, depending on which segments are treated as exons during splicing
- This is called **alternative RNA splicing**
- Consequently, the number of different proteins an organism can produce is much greater than its number of genes

- Proteins often have a modular architecture consisting of discrete regions called **domains**
- In many cases, different exons code for the different domains in a protein
- Exon shuffling may result in the evolution of new proteins

Figure 17.13



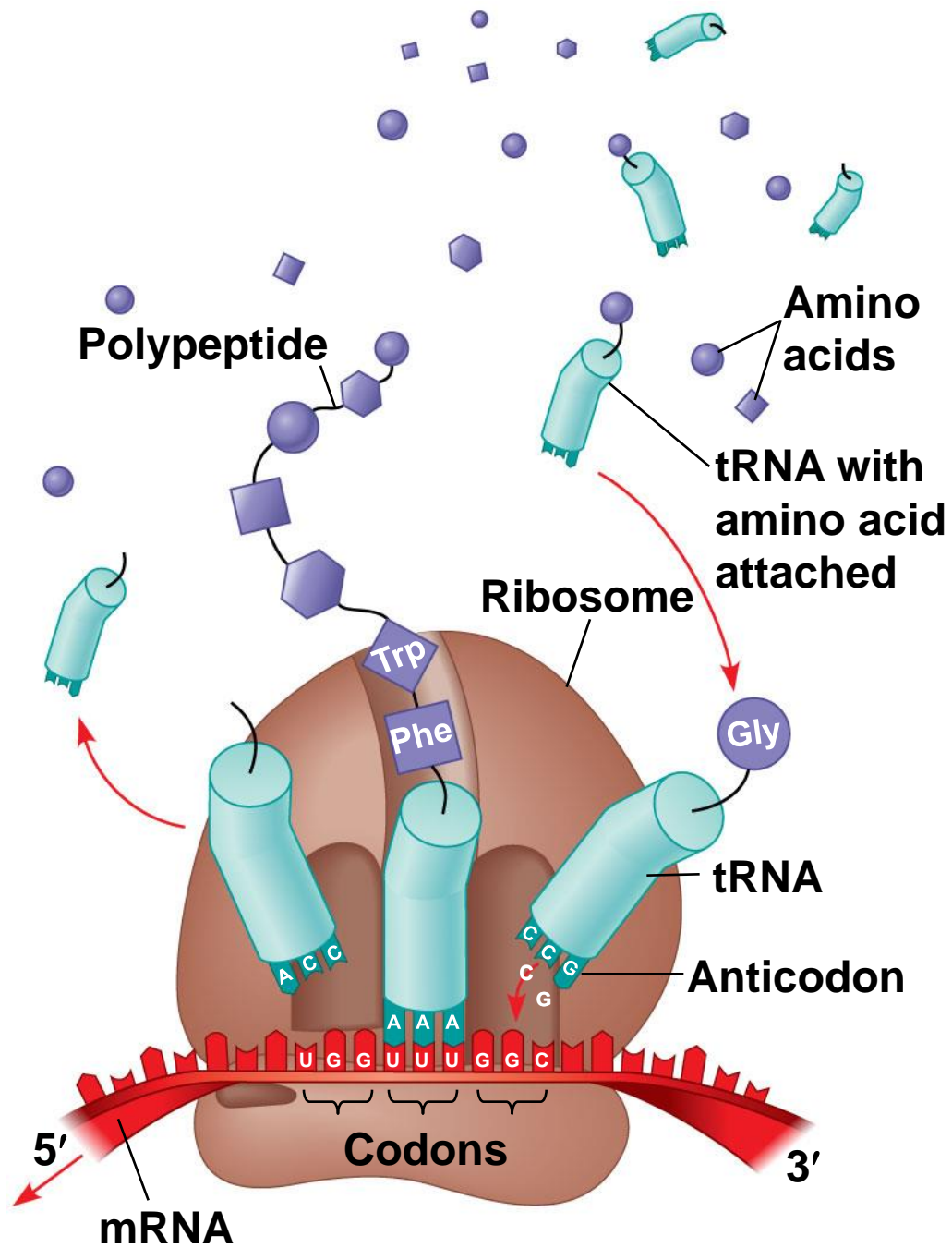
Concept 17.4: Translation is the RNA-directed synthesis of a polypeptide: *a closer look*

- Genetic information flows from mRNA to protein through the process of translation

Molecular Components of Translation

- A cell translates an mRNA message into protein with the help of **transfer RNA (tRNA)**
- tRNAs transfer amino acids to the growing polypeptide in a ribosome
- Translation is a complex process in terms of its biochemistry and mechanics

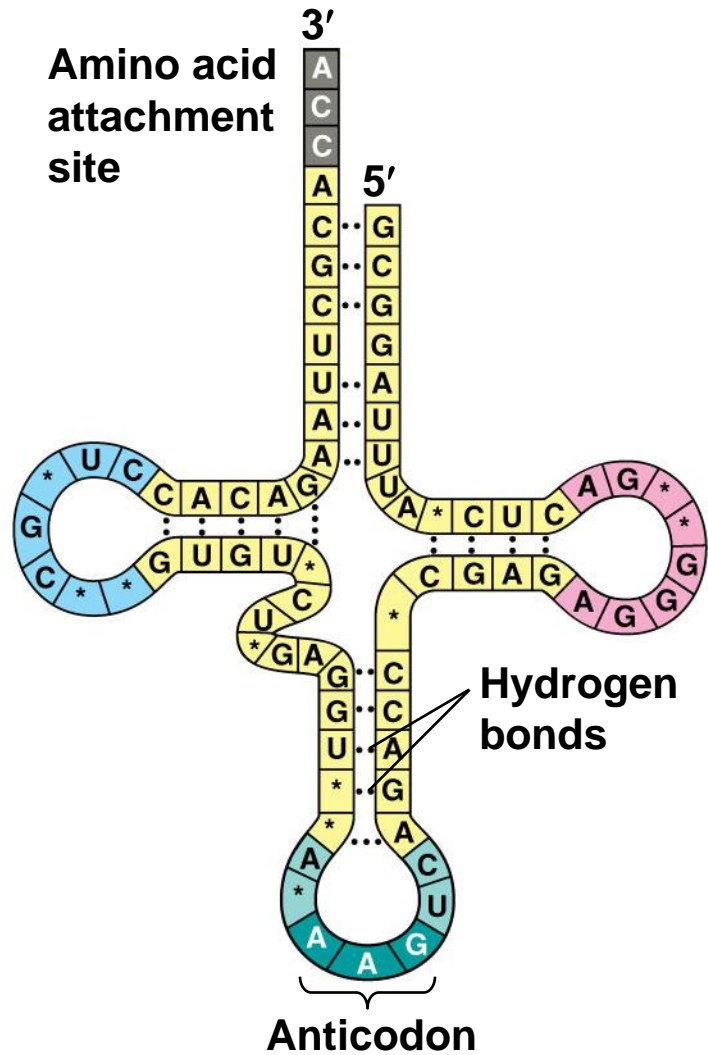
Figure 17.14



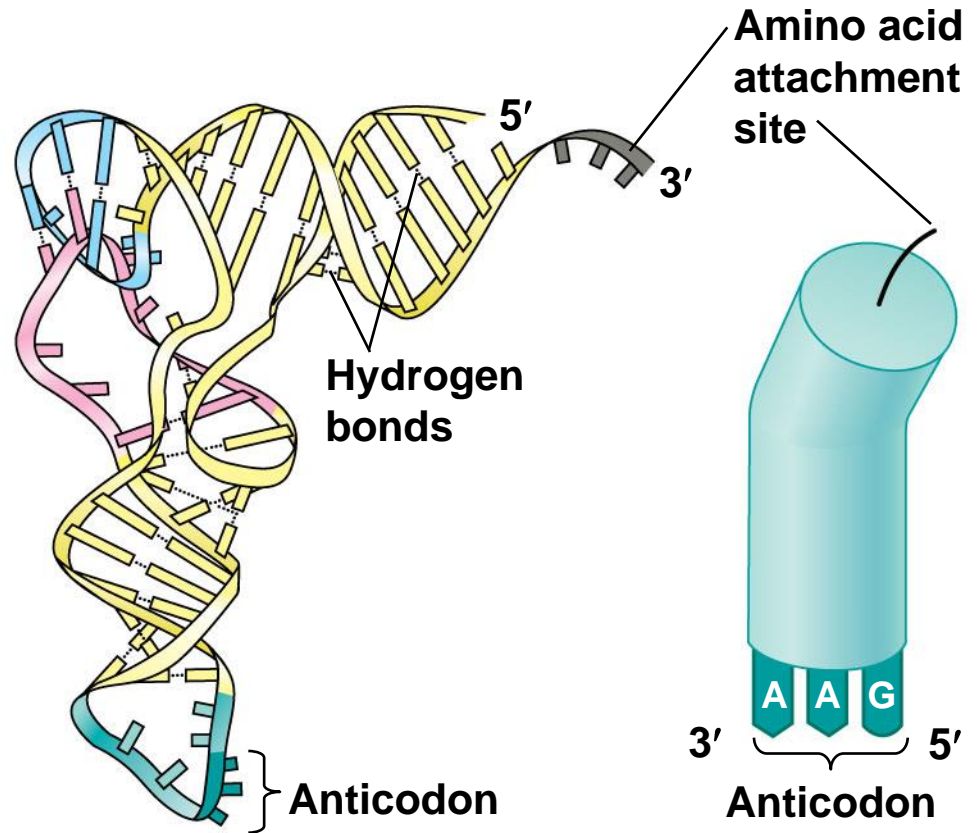
The Structure and Function of Transfer RNA

- Molecules of tRNA are not identical
 - Each carries a specific amino acid on one end
 - Each has an **anticodon** on the other end; the anticodon base-pairs with a complementary codon on mRNA

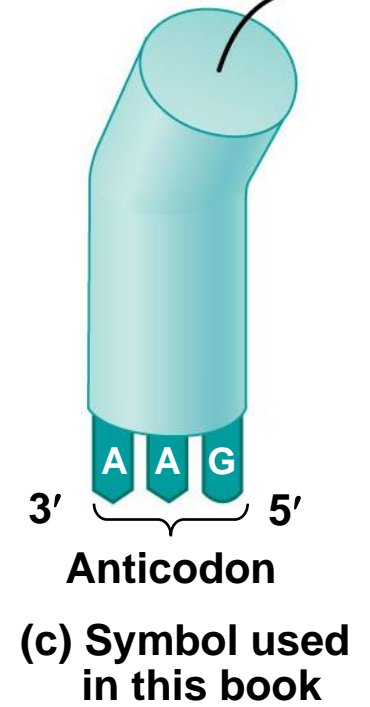
- A tRNA molecule consists of a single RNA strand that is only about 80 nucleotides long
- Flattened into one plane to reveal its base pairing, a tRNA molecule looks like a cloverleaf



(a) Two-dimensional structure



(b) Three-dimensional structure

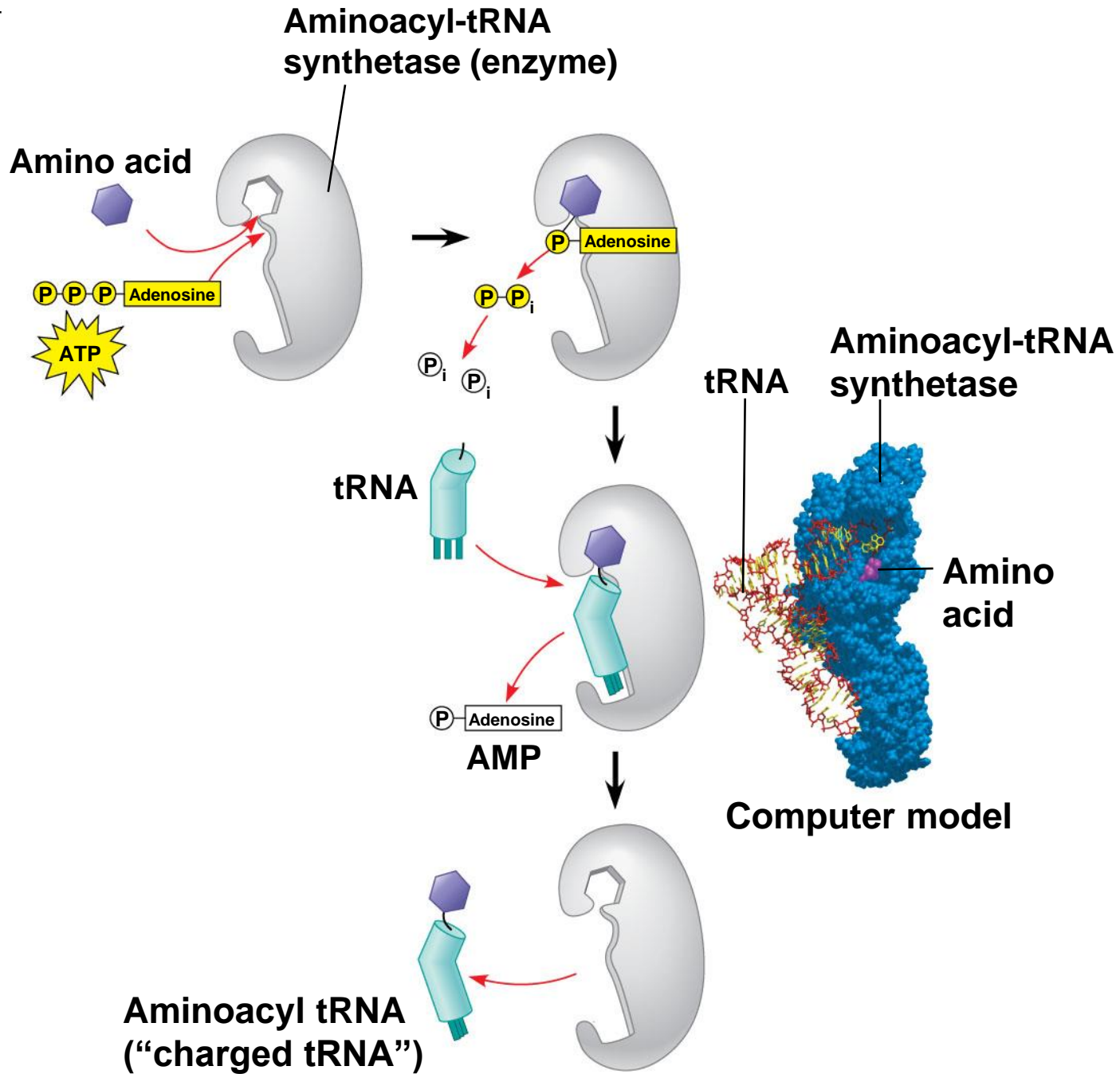


(c) Symbol used in this book

- Because of hydrogen bonds, tRNA actually twists and folds into a three-dimensional molecule
- tRNA is roughly L-shaped

- Accurate translation requires two steps
 - First: a correct match between a tRNA and an amino acid, done by the enzyme **aminoacyl-tRNA synthetase**
 - Second: a correct match between the tRNA anticodon and an mRNA codon
- Flexible pairing at the third base of a codon is called **wobble** and allows some tRNAs to bind to more than one codon

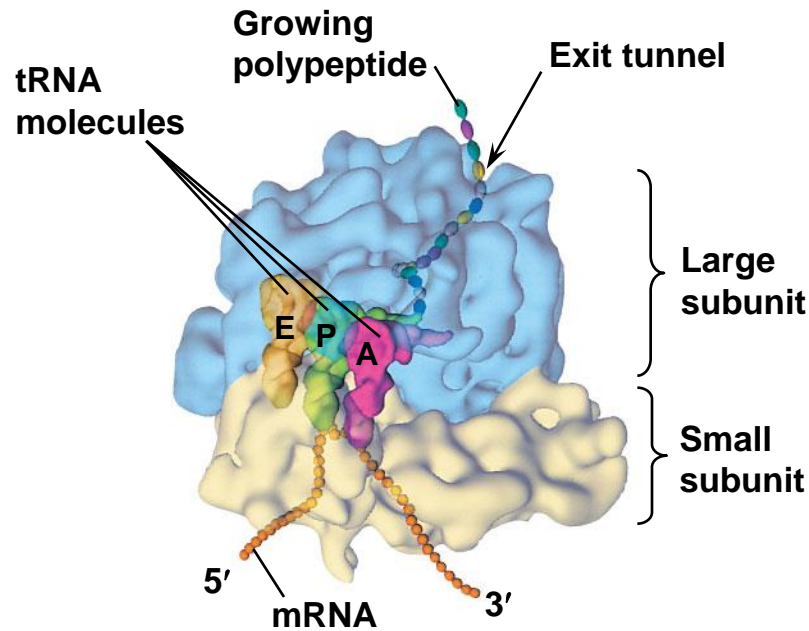
Figure 17.16-4



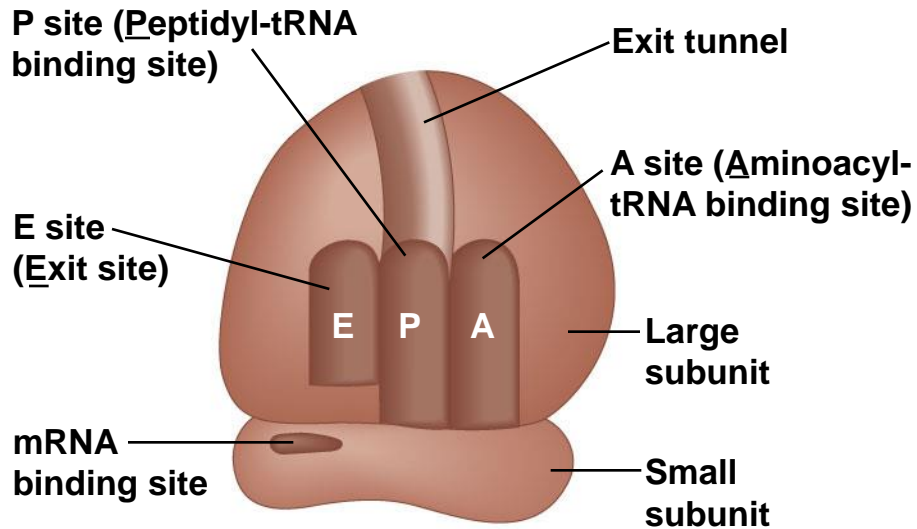
Ribosomes

- Ribosomes facilitate specific coupling of tRNA anticodons with mRNA codons in protein synthesis
- The two ribosomal subunits (large and small) are made of proteins and **ribosomal RNA (rRNA)**
- Bacterial and eukaryotic ribosomes are somewhat similar but have significant differences: some antibiotic drugs specifically target bacterial ribosomes without harming eukaryotic ribosomes

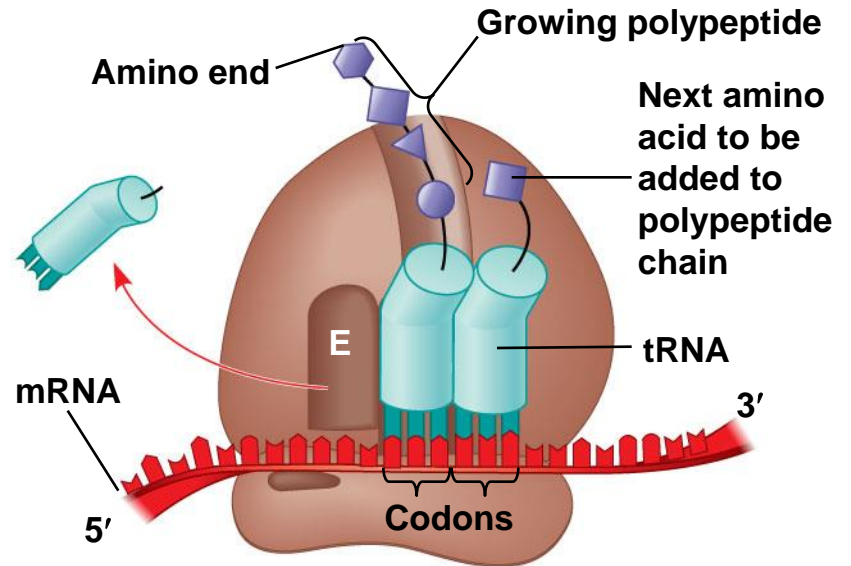
Figure 17.17



(a) Computer model of functioning ribosome



(b) Schematic model showing binding sites



(c) Schematic model with mRNA and tRNA

- A ribosome has three binding sites for tRNA
 - The **P site** holds the tRNA that carries the growing polypeptide chain
 - The **A site** holds the tRNA that carries the next amino acid to be added to the chain
 - The **E site** is the exit site, where discharged tRNAs leave the ribosome

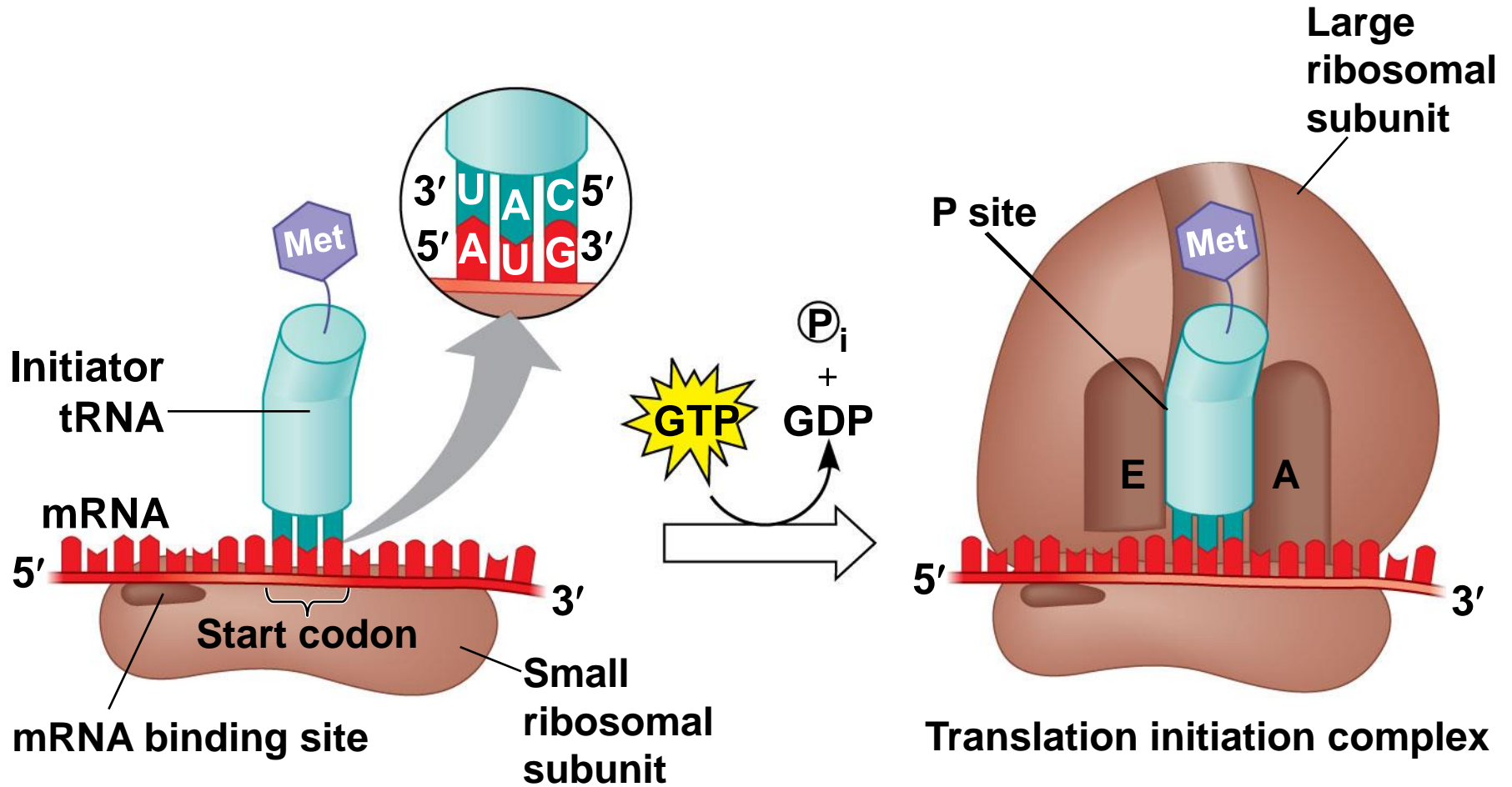
Building a Polypeptide

- The three stages of translation
 - Initiation
 - Elongation
 - Termination
- All three stages require protein “factors” that aid in the translation process

Ribosome Association and Initiation of Translation

- The initiation stage of translation brings together mRNA, a tRNA with the first amino acid, and the two ribosomal subunits
- First, a small ribosomal subunit binds with mRNA and a special initiator tRNA
- Then the small subunit moves along the mRNA until it reaches the start codon (AUG)
- Proteins called initiation factors bring in the large subunit that completes the translation initiation complex

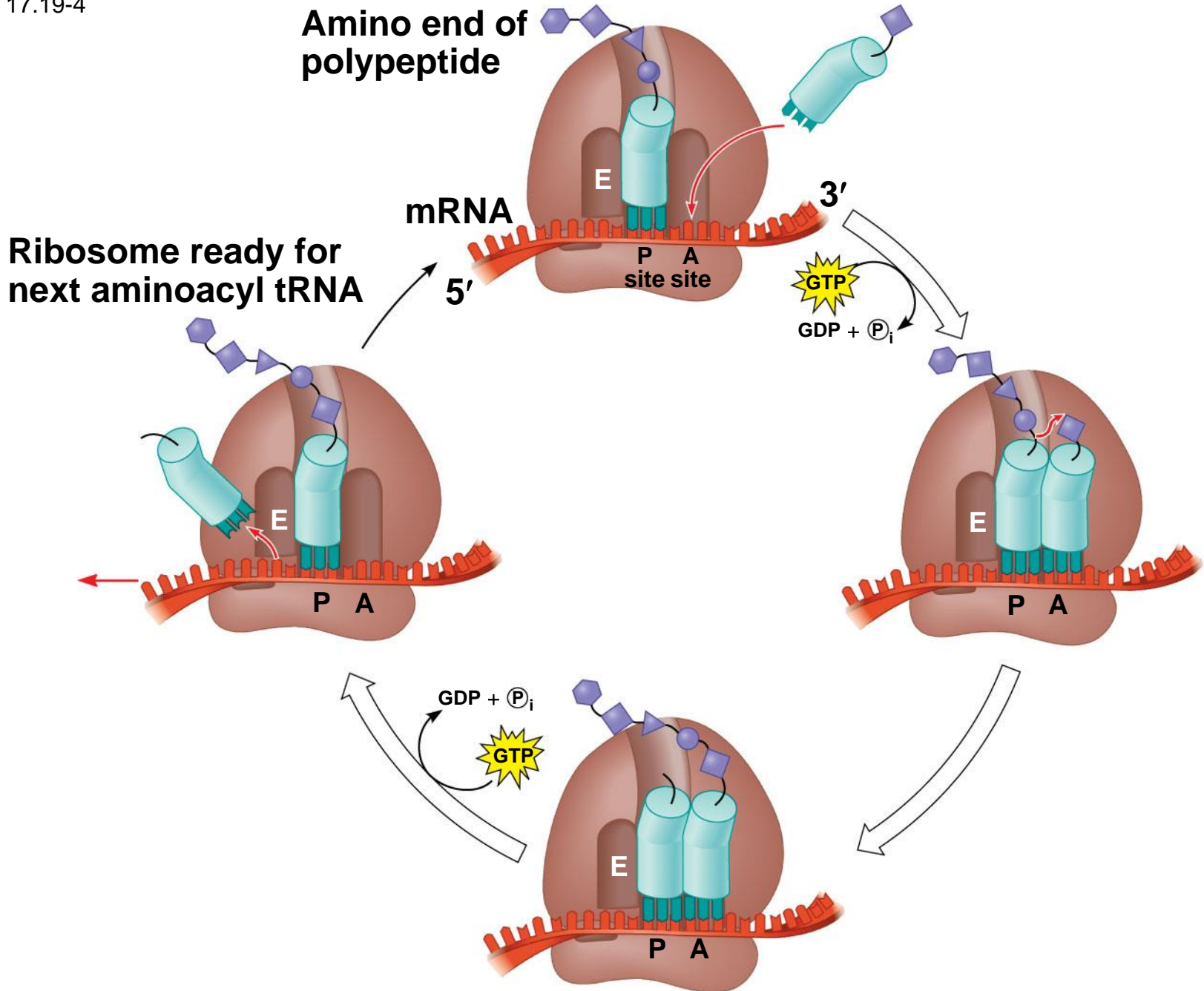
Figure 17.18



Elongation of the Polypeptide Chain

- During the elongation stage, amino acids are added one by one to the preceding amino acid at the C-terminus of the growing chain
- Each addition involves proteins called elongation factors and occurs in three steps: codon recognition, peptide bond formation, and translocation
- Translation proceeds along the mRNA in a 5' to 3' direction

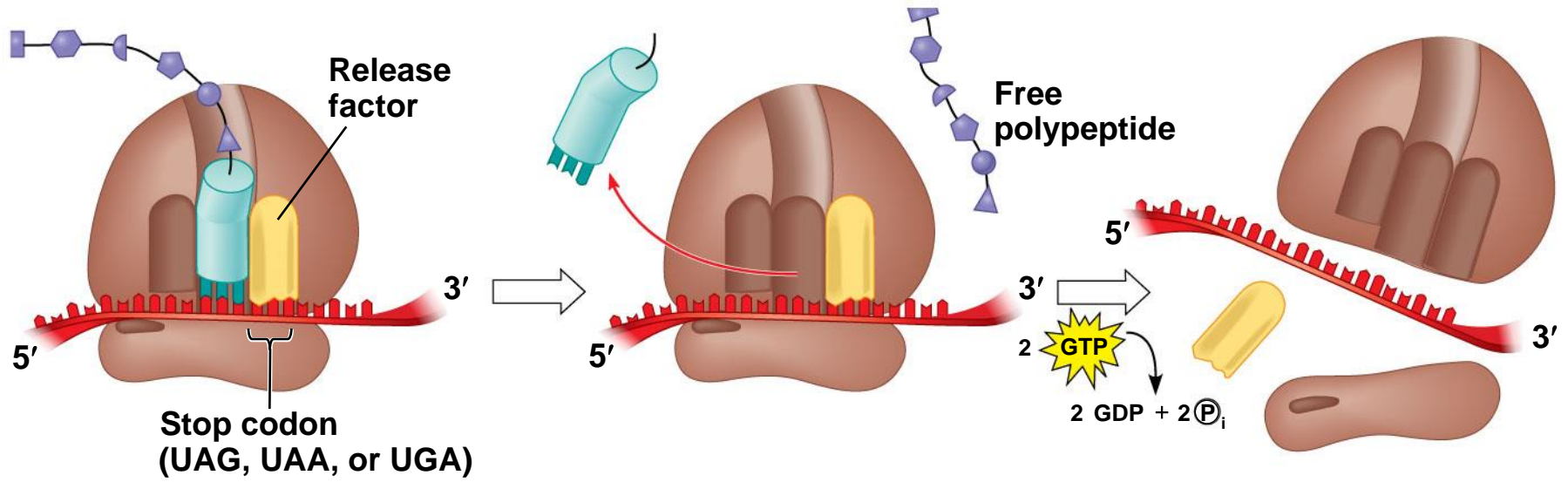
Figure 17.19-4



Termination of Translation

- Termination occurs when a stop codon in the mRNA reaches the A site of the ribosome
- The A site accepts a protein called a release factor
- The release factor causes the addition of a water molecule instead of an amino acid
- This reaction releases the polypeptide, and the translation assembly then comes apart

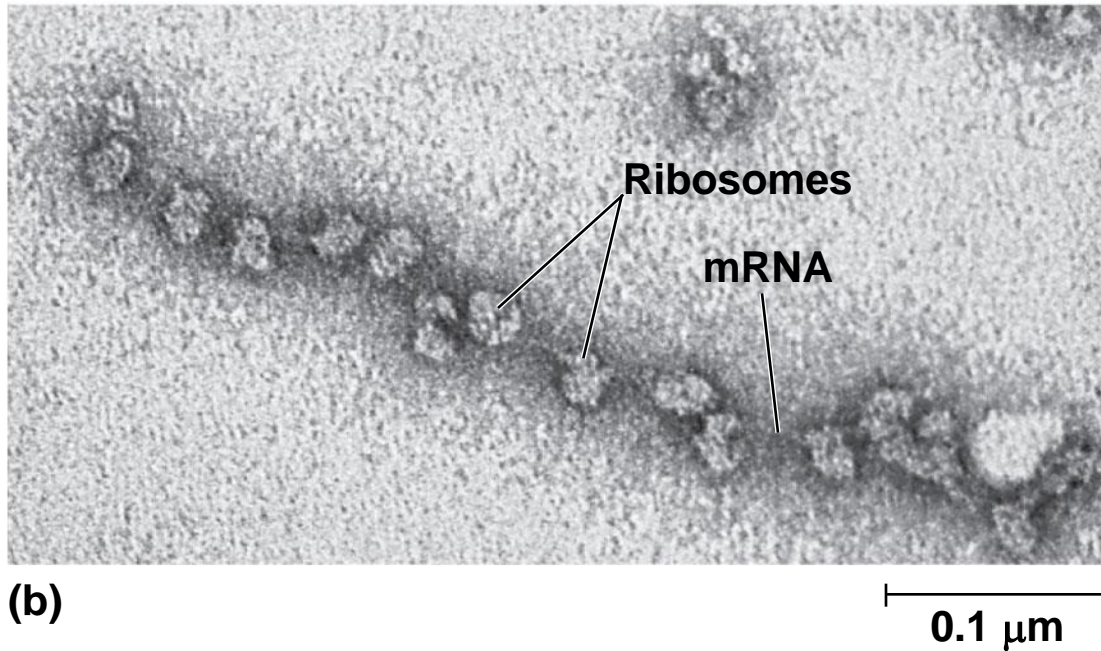
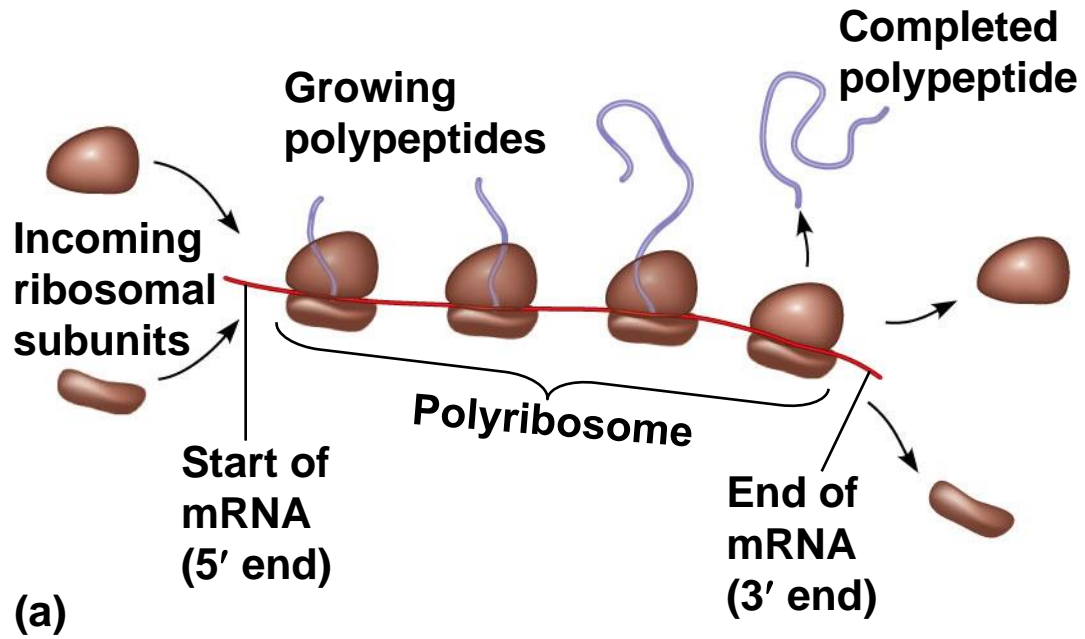
Figure 17.20-3



Polyribosomes

- A number of ribosomes can translate a single mRNA simultaneously, forming a **polyribosome** (or **polysome**)
- Polyribosomes enable a cell to make many copies of a polypeptide very quickly

Figure 17.21



Completing and Targeting the Functional Protein

- Often translation is not sufficient to make a functional protein
- Polypeptide chains are modified after translation or targeted to specific sites in the cell

Protein Folding and Post-Translational Modifications

- During and after synthesis, a polypeptide chain spontaneously coils and folds into its three-dimensional shape
- Proteins may also require post-translational modifications before doing their job
- Some polypeptides are activated by enzymes that cleave them
- Other polypeptides come together to form the subunits of a protein

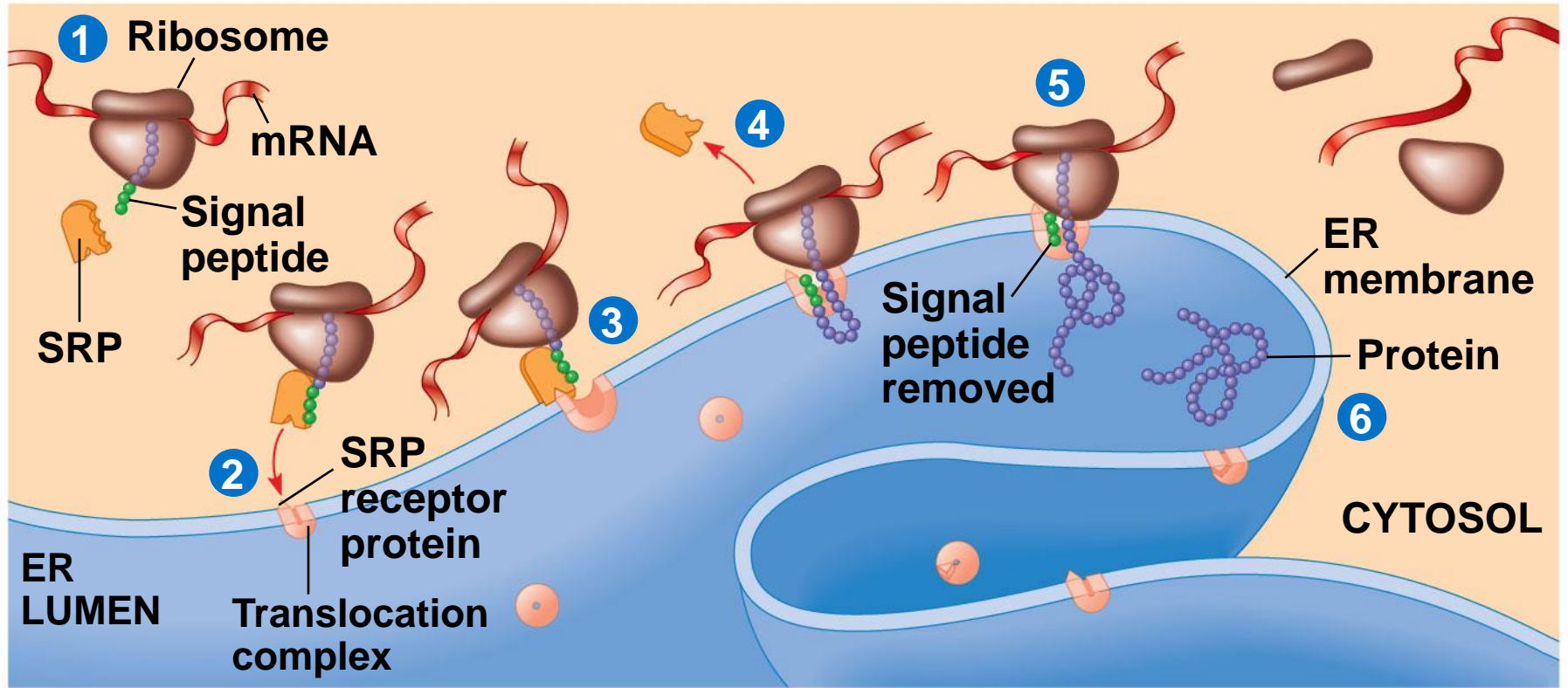
Targeting Polypeptides to Specific Locations

- Two populations of ribosomes are evident in cells: free ribosomes (in the cytosol) and bound ribosomes (attached to the ER)
- Free ribosomes mostly synthesize proteins that function in the cytosol
- Bound ribosomes make proteins of the endomembrane system and proteins that are secreted from the cell
- Ribosomes are identical and can switch from free to bound

- Polypeptide synthesis always begins in the cytosol
- Synthesis finishes in the cytosol *unless* the polypeptide signals the ribosome to attach to the ER
- Polypeptides destined for the ER or for secretion are marked by a **signal peptide**

- A **signal-recognition particle (SRP)** binds to the signal peptide
- The SRP brings the signal peptide and its ribosome to the ER

Figure 17.22



Concept 17.5: Mutations of one or a few nucleotides can affect protein structure and function

- **Mutations** are changes in the genetic material of a cell or virus
- **Point mutations** are chemical changes in just one base pair of a gene
- The change of a single nucleotide in a DNA template strand can lead to the production of an abnormal protein

Wild-type hemoglobin

Wild-type hemoglobin DNA



mRNA



Normal hemoglobin



Sickle-cell hemoglobin

Mutant hemoglobin DNA



mRNA



Sickle-cell hemoglobin



Types of Small-Scale Mutations

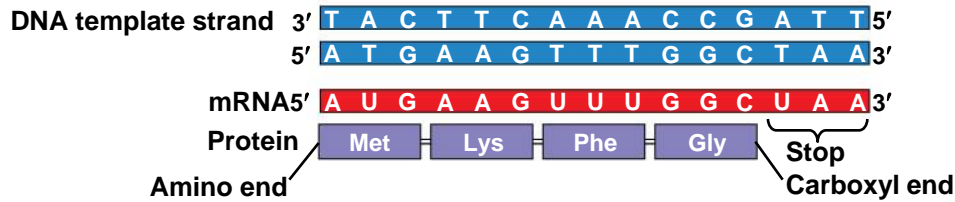
- Point mutations within a gene can be divided into two general categories
 - Nucleotide-pair substitutions
 - One or more nucleotide-pair insertions or deletions

Substitutions

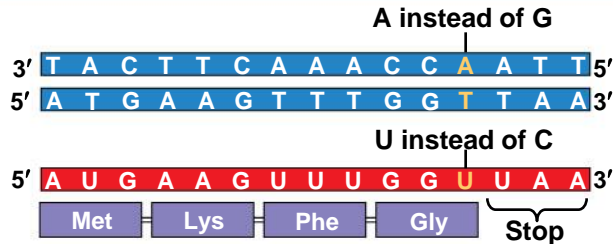
- A **nucleotide-pair substitution** replaces one nucleotide and its partner with another pair of nucleotides
- **Silent mutations** have no effect on the amino acid produced by a codon because of redundancy in the genetic code
- **Missense mutations** still code for an amino acid, but not the correct amino acid
- **Nonsense mutations** change an amino acid codon into a stop codon, nearly always leading to a nonfunctional protein

Figure 17.24

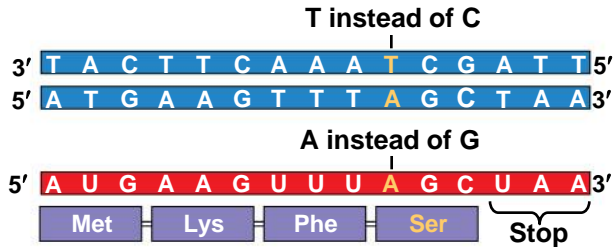
Wild type



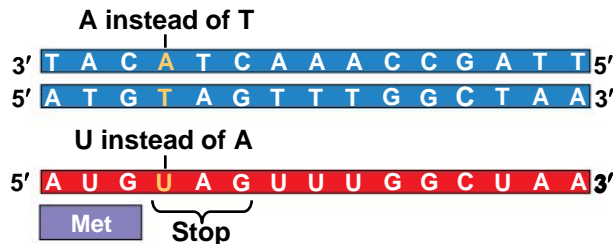
(a) Nucleotide-pair substitution



Silent (no effect on amino acid sequence)

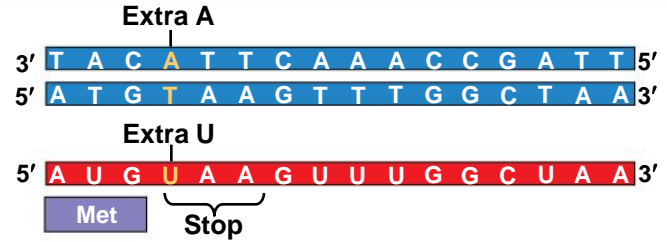


Missense

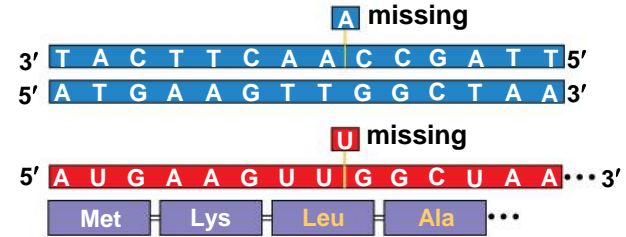


Nonsense

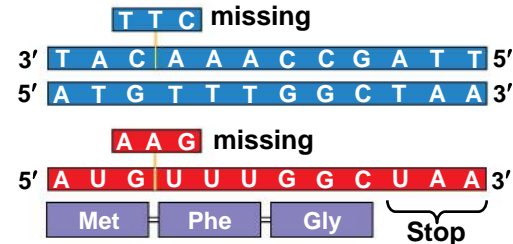
(b) Nucleotide-pair insertion or deletion



Frameshift causing immediate nonsense (1 nucleotide-pair insertion)



Frameshift causing extensive missense (1 nucleotide-pair deletion)



No frameshift, but one amino acid missing (3 nucleotide-pair deletion)

Insertions and Deletions

- **Insertions** and **deletions** are additions or losses of nucleotide pairs in a gene
- These mutations have a disastrous effect on the resulting protein more often than substitutions do
- Insertion or deletion of nucleotides may alter the reading frame, producing a **frameshift mutation**

Mutagens

- Spontaneous mutations can occur during DNA replication, recombination, or repair
- **Mutagens** are physical or chemical agents that can cause mutations

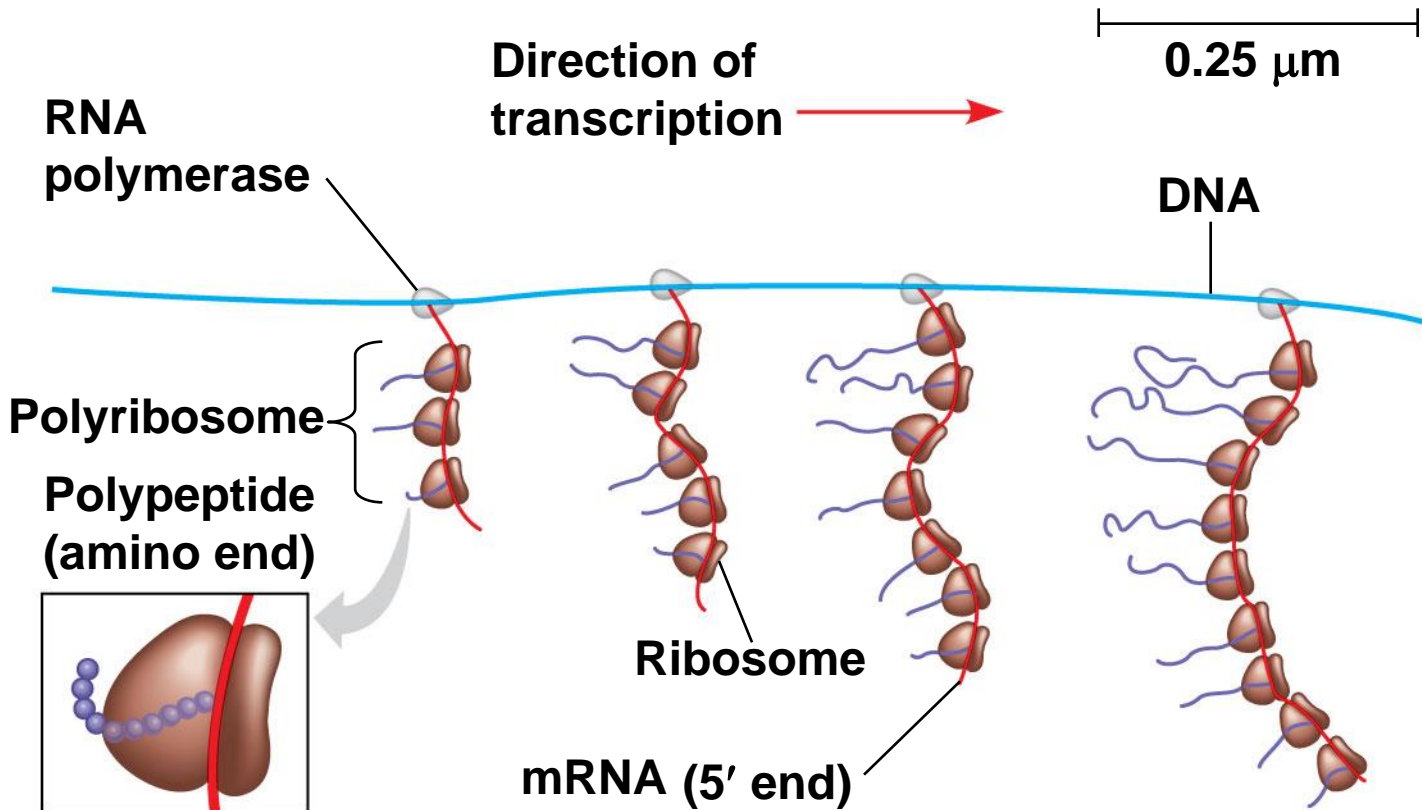
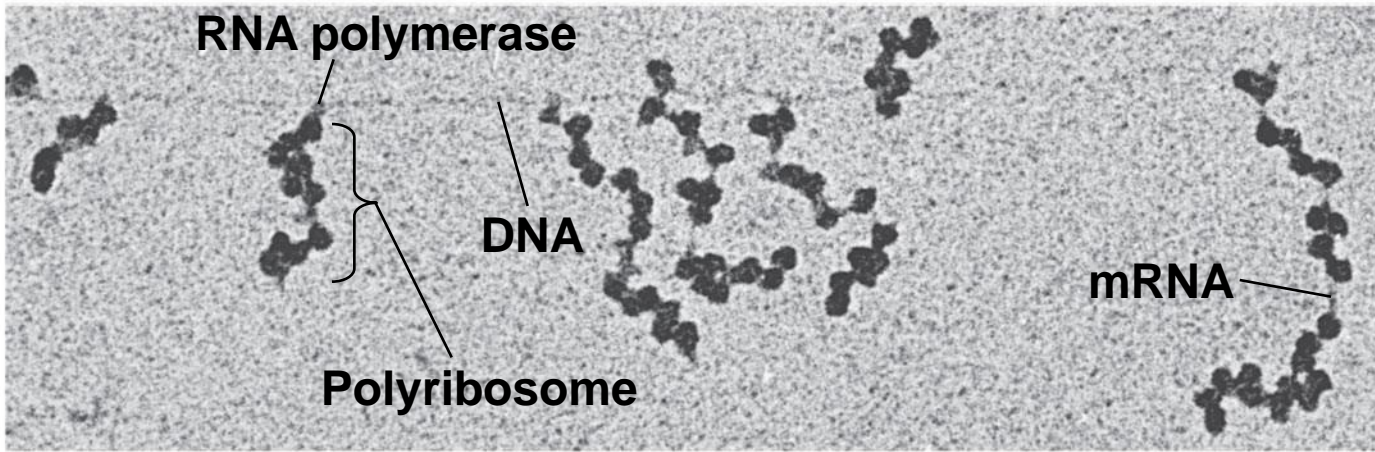
Concept 17.6: While gene expression differs among the domains of life, the concept of a gene is universal

- Archaea are prokaryotes, but share many features of gene expression with eukaryotes

Comparing Gene Expression in Bacteria, Archaea, and Eukarya

- Bacteria and eukarya differ in their RNA polymerases, termination of transcription, and ribosomes; archaea tend to resemble eukarya in these respects
- Bacteria can simultaneously transcribe and translate the same gene
- In eukarya, transcription and translation are separated by the nuclear envelope
- In archaea, transcription and translation are likely coupled

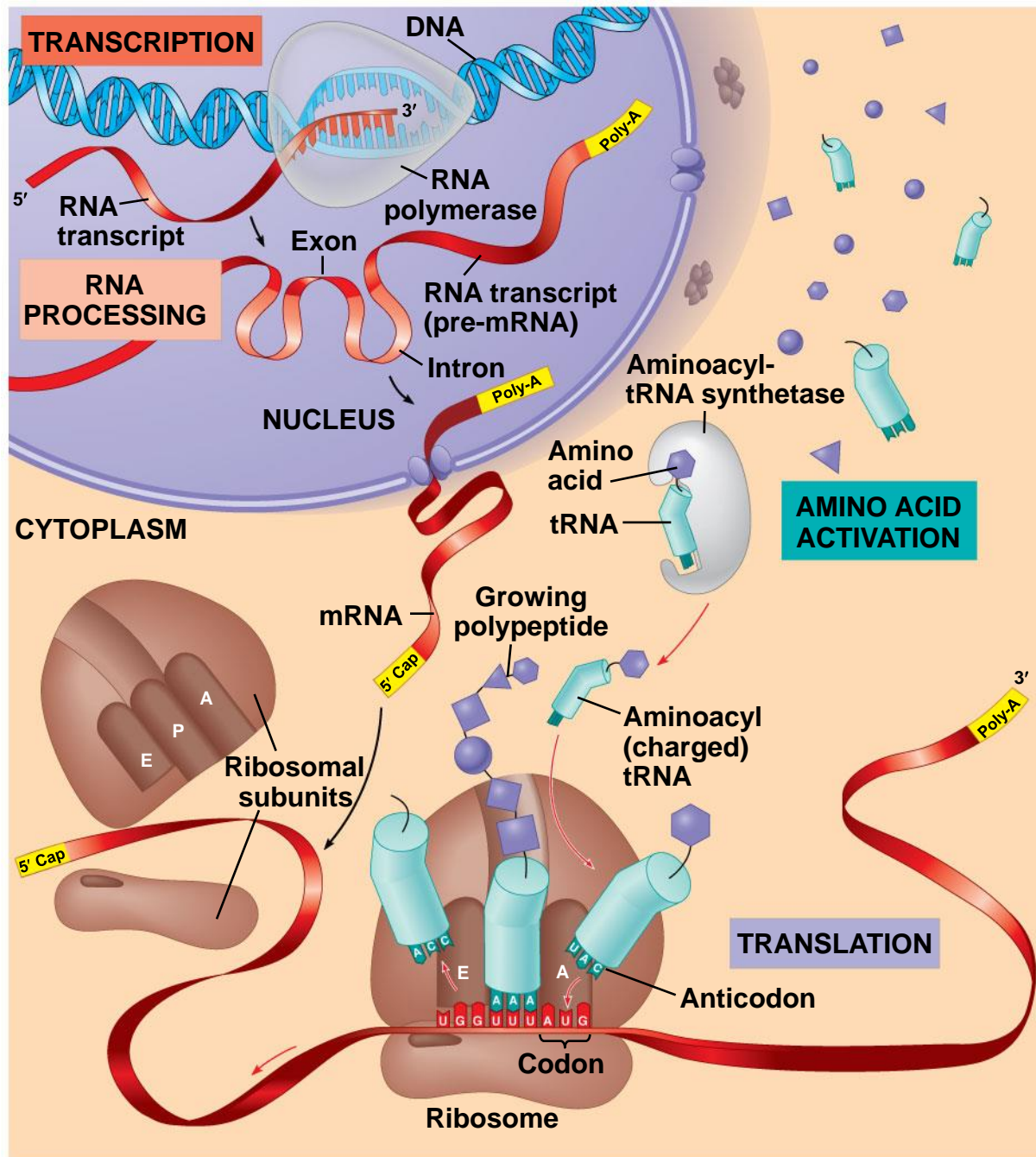
Figure 17.25



What Is a Gene? *Revisiting the Question*

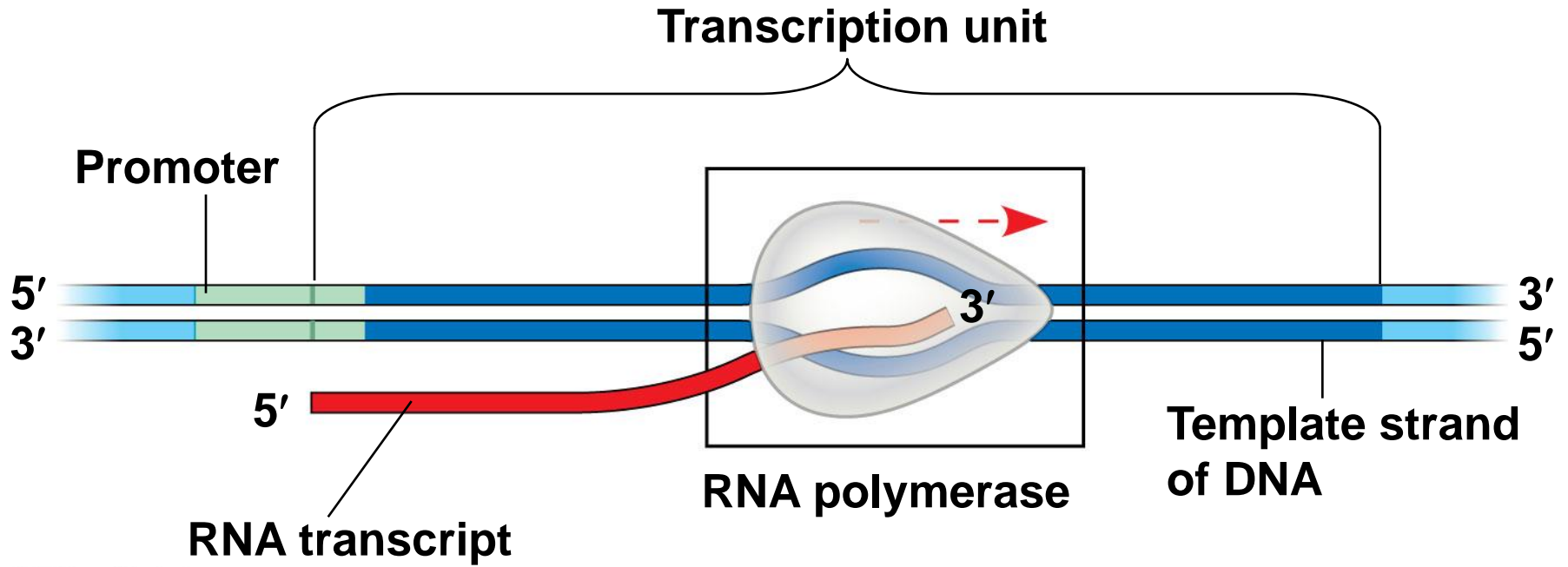
- The idea of the gene has evolved through the history of genetics
- We have considered a gene as
 - A discrete unit of inheritance
 - A region of specific nucleotide sequence in a chromosome
 - A DNA sequence that codes for a specific polypeptide chain

Figure 17.26



- In summary, a gene can be defined as a region of DNA that can be expressed to produce a final functional product, either a polypeptide or an RNA molecule

Figure 17.UN02



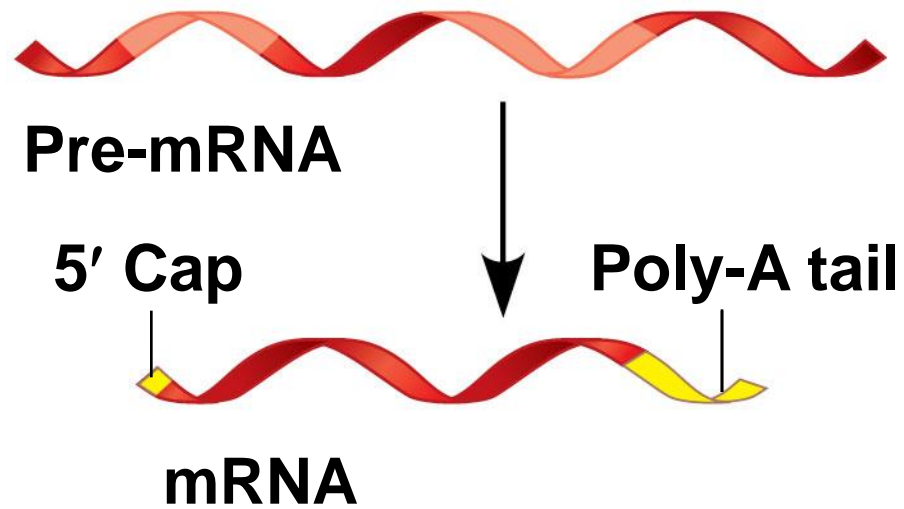


Figure 17.UN04

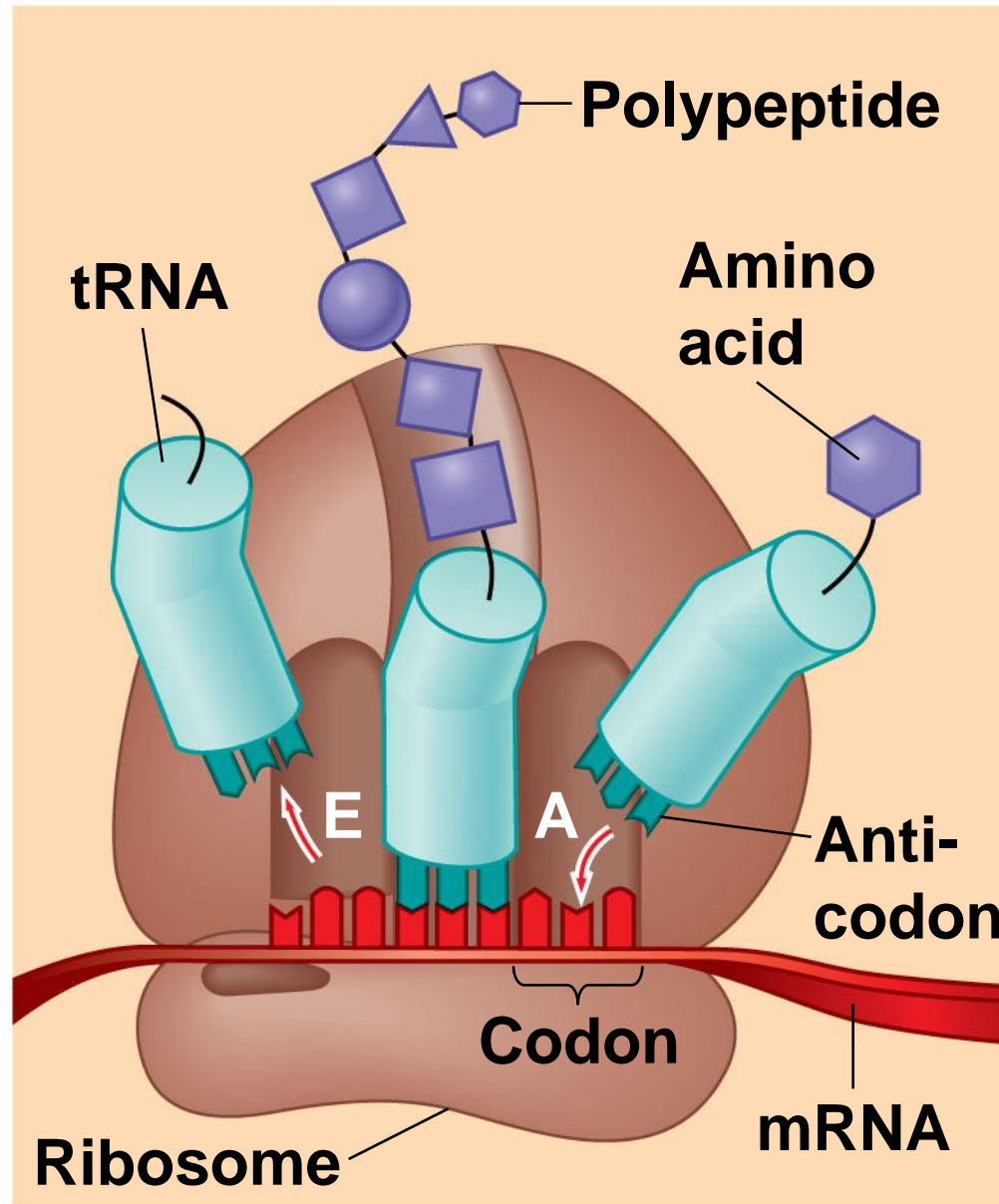


Figure 17.UN05

Type of RNA	Functions
Messenger RNA (mRNA)	
Transfer RNA (tRNA)	
	Plays catalytic (ribozyme) roles and structural roles in ribosomes
Primary transcript	
Small nuclear RNA (snRNA)	

Template sequence
(from problem):

3'-TTCAGTCGT-5'

Nontemplate sequence:

5'-AAGTCAGCA-3'

mRNA sequence:

5'-AAGUCAGCA-3'

"Template sequence" (from nontemplate sequence in problem,
written 3' → 5'):

3'-ACGACTGAA-5'

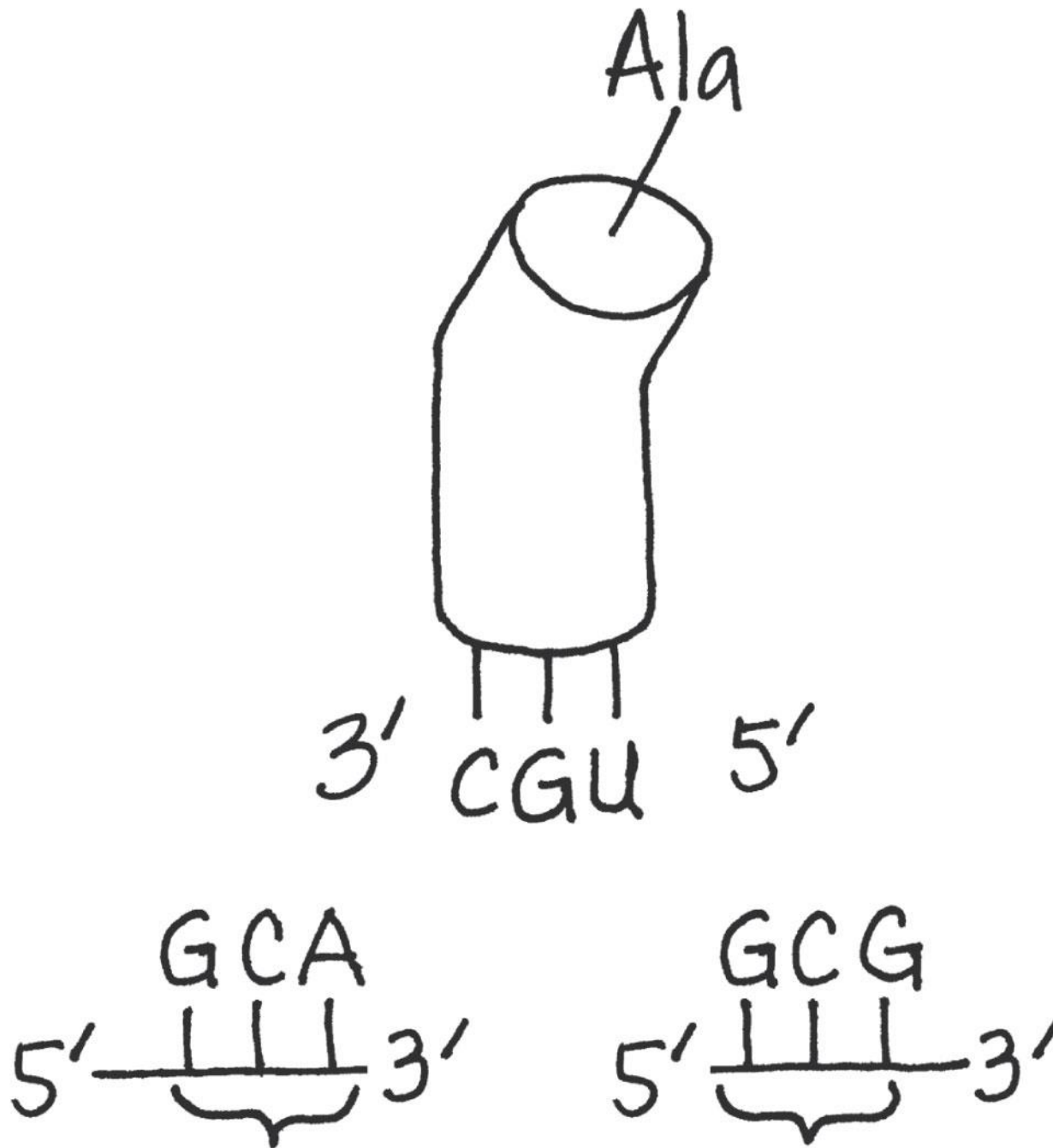
mRNA sequence:

5'-UGCUGACUU-3'

Translated:

Cys-STOP-Leu

Figure 17.UN06c



Normal DNA sequence
(template strand is on top):

3'-TACTTGTCCGATATC-5'
5'-ATGAACAGGCTATAG-3'

mRNA sequence:

5'-AUGAACAGGCUAUAG-3'

Amino acid sequence:

Met-Asn-Arg-Leu-STOP

Mutated DNA sequence
(template strand is on top):

3'-TACTTGTCCAATATC-5'
5'-ATGAACAGGTTATAG-3'

mRNA sequence:

5'-AUGAACAGGUUAUAG-3'

Amino acid sequence:

Met-Asn-Arg-Leu-STOP

Figure 17.UN10

Type of RNA	Functions
Messenger RNA (mRNA)	Carries information specifying amino acid sequences of proteins from DNA to ribosomes
Transfer RNA (tRNA)	Serves as translator molecule in protein synthesis; translates mRNA codons into amino acids
Ribosomal RNA (rRNA)	Plays catalytic (ribozyme) roles and structural roles in ribosomes
Primary transcript	Is a precursor to mRNA, rRNA, or tRNA, before being processed; some intron RNA acts as a ribozyme, catalyzing its own splicing
Small nuclear RNA (snRNA)	Plays structural and catalytic roles in spliceosomes, the complexes of protein and RNA that splice pre-mRNA