Chapter 16 TRANSCRIPTION AND TRANSLATION

DNA base sequence to protein

SUMMARY

- Review of genes, RNA and the “code
- Transcription
  - Initiation
  - Elongation and termination
- Translation
  - Initiation
  - Elongation and termination
- Mutations

Genes - Review

- Genes are particular base sequences on DNA
- Genes code for proteins that produce the characteristics (phenotype) of an organism?
- Proteins are made in the cytoplasm on ribosomes.
- How does DNA direct protein synthesis?

RNA-THE PROTEIN ASSEMBLY LINE AND WORKERS

RNA - 3 types

rRNA = ribosomal RNA; combined with proteins forms ribosomes.

tRNA = transfer RNA; transports amino acids to ribosomes for protein assembly.

mRNA = messenger RNA; brings "code" (instructions) for the protein from the nucleus to the cytoplasm.

General Scheme – 2 Steps

The DNA code for a protein is copied to mRNA = transcription.

mRNA code used to assemble protein = translation.

The Code

- A specific order of nucleotides or bases on the DNA.
- Occurs in blocks of 3 bases = <u>codons</u> that specify which amino acid goes where in a protein.
- 1 codon = one amino acid
- The code is “universal”, i.e. the codons specify the same amino acids in all organisms, pretty much.

Transcription

Production of a mRNA "copy" of the DNA sequence of a certain gene.

Enzyme = RNA polymerase; 1 in prokaryotes, 3 in eukaryotes.

The base sequence on 1 DNA strand is used as a template = "template strand". The other strand is the "non-template strand", fig 16.1.

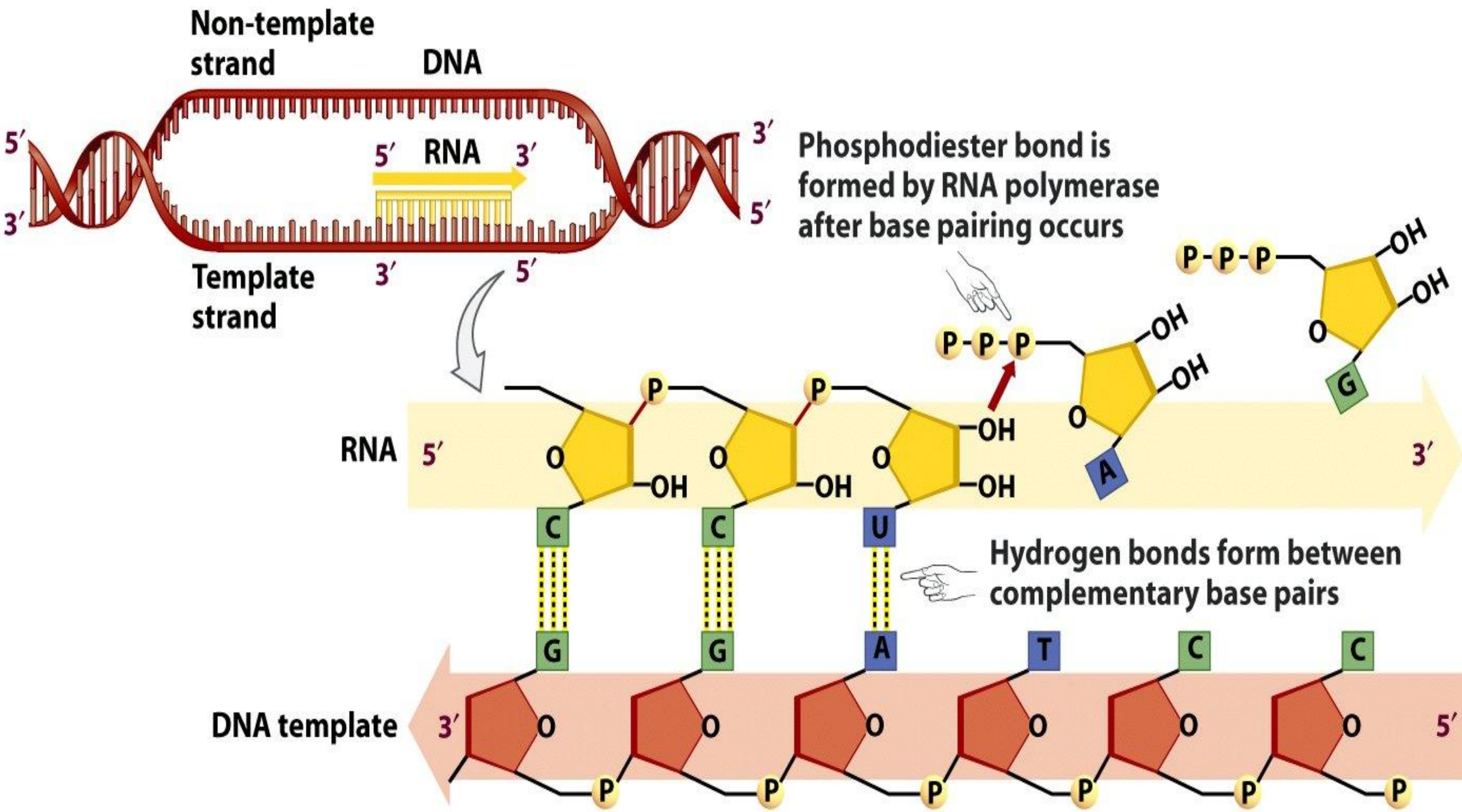


Figure 16-1 Biological Science, 2/e

Transcription

- Sequence in prokaryotes, fig 16.3.
  - Initiation phase
    - RNA polymerase bind a Sigma protein that in turn binds to the promoter, upstream from the start site for a particular gene on DNA.
    - Sigma opens up a portion of the DNA helix and RNA polymerase begins transcription.

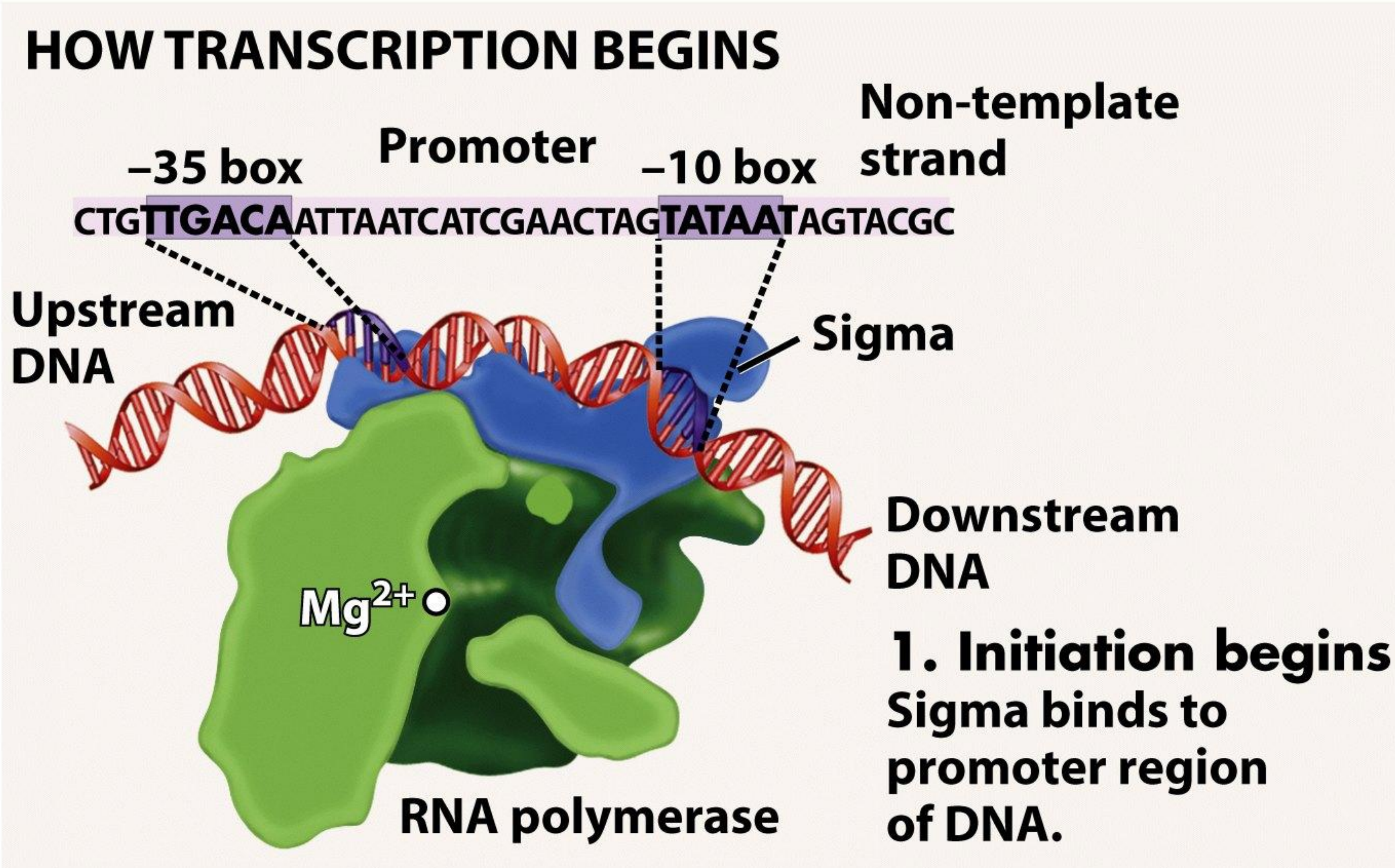
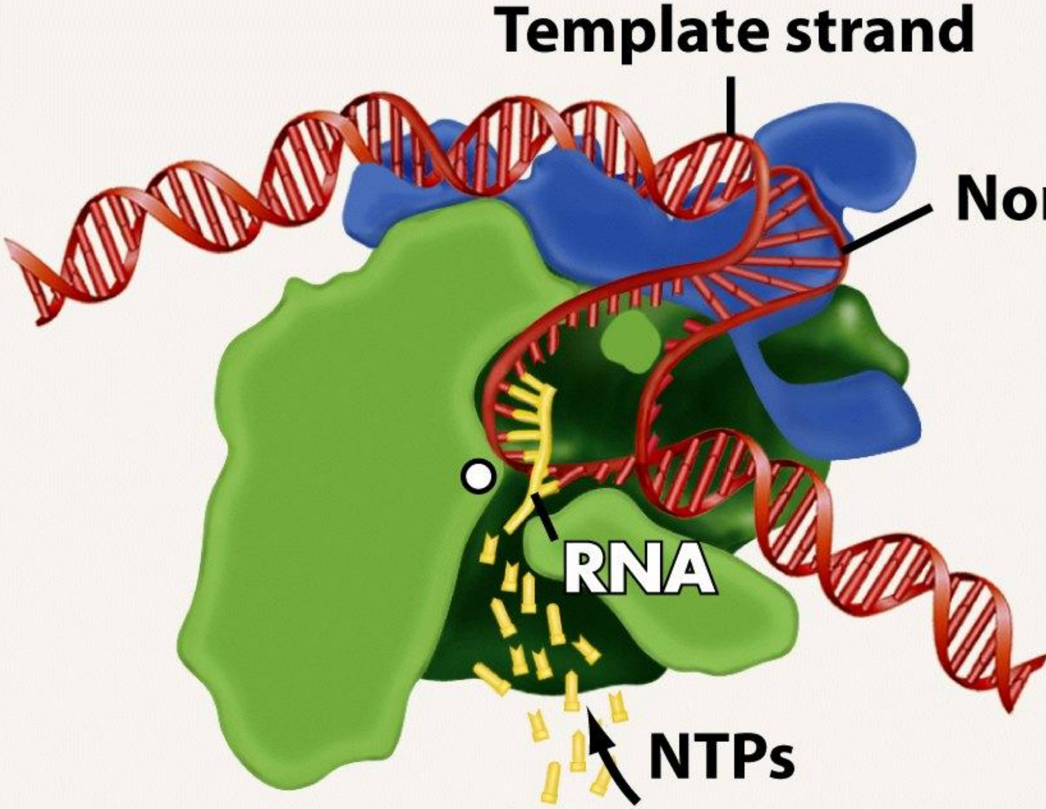


Figure 16-3 part 1 Biological Science, 2/e



2. Initiation continues
Sigma opens the DNA helix; transcription begins.

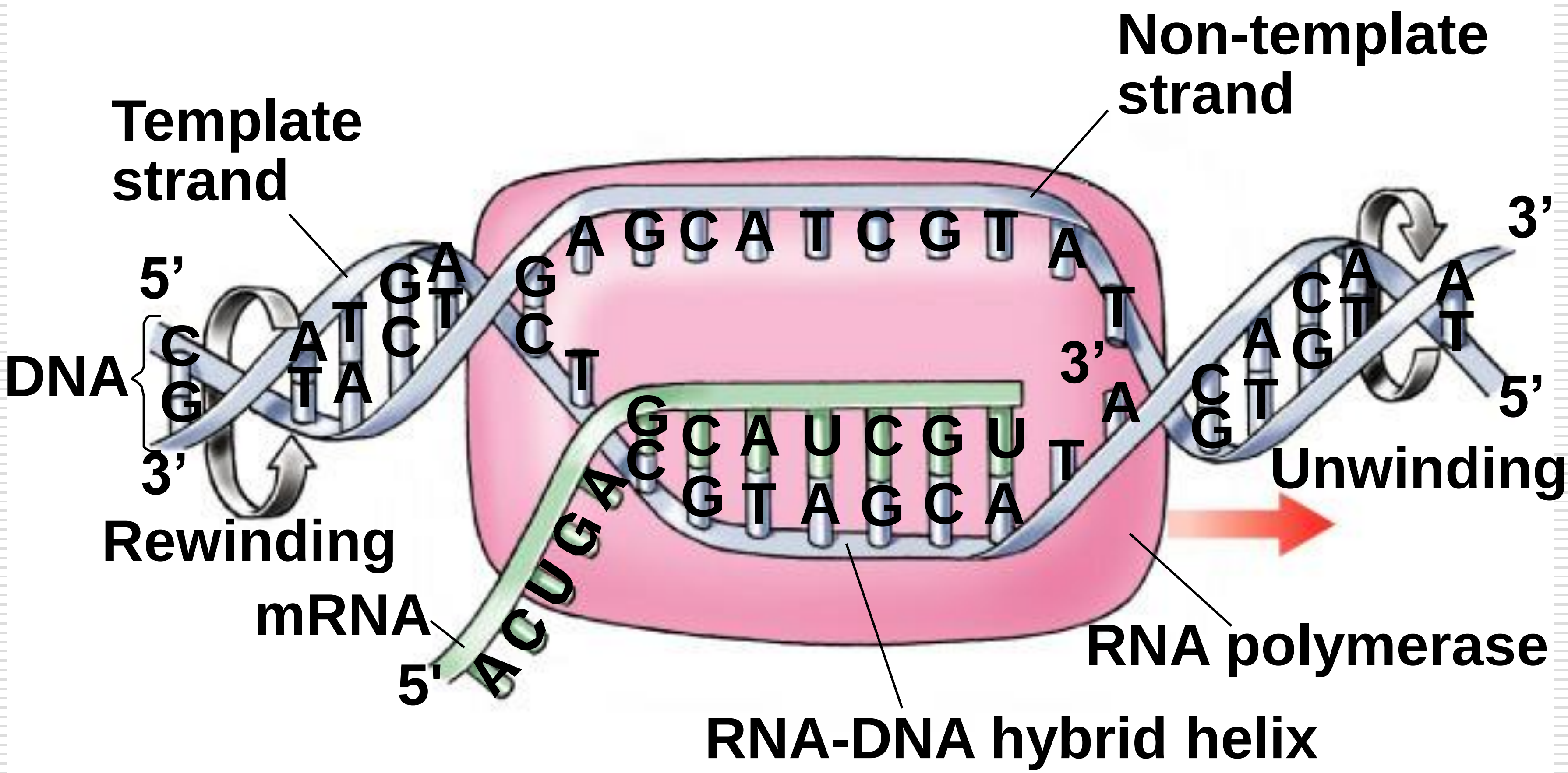
Figure 16-3 part 2 Biological Science, 2/e

Transcription

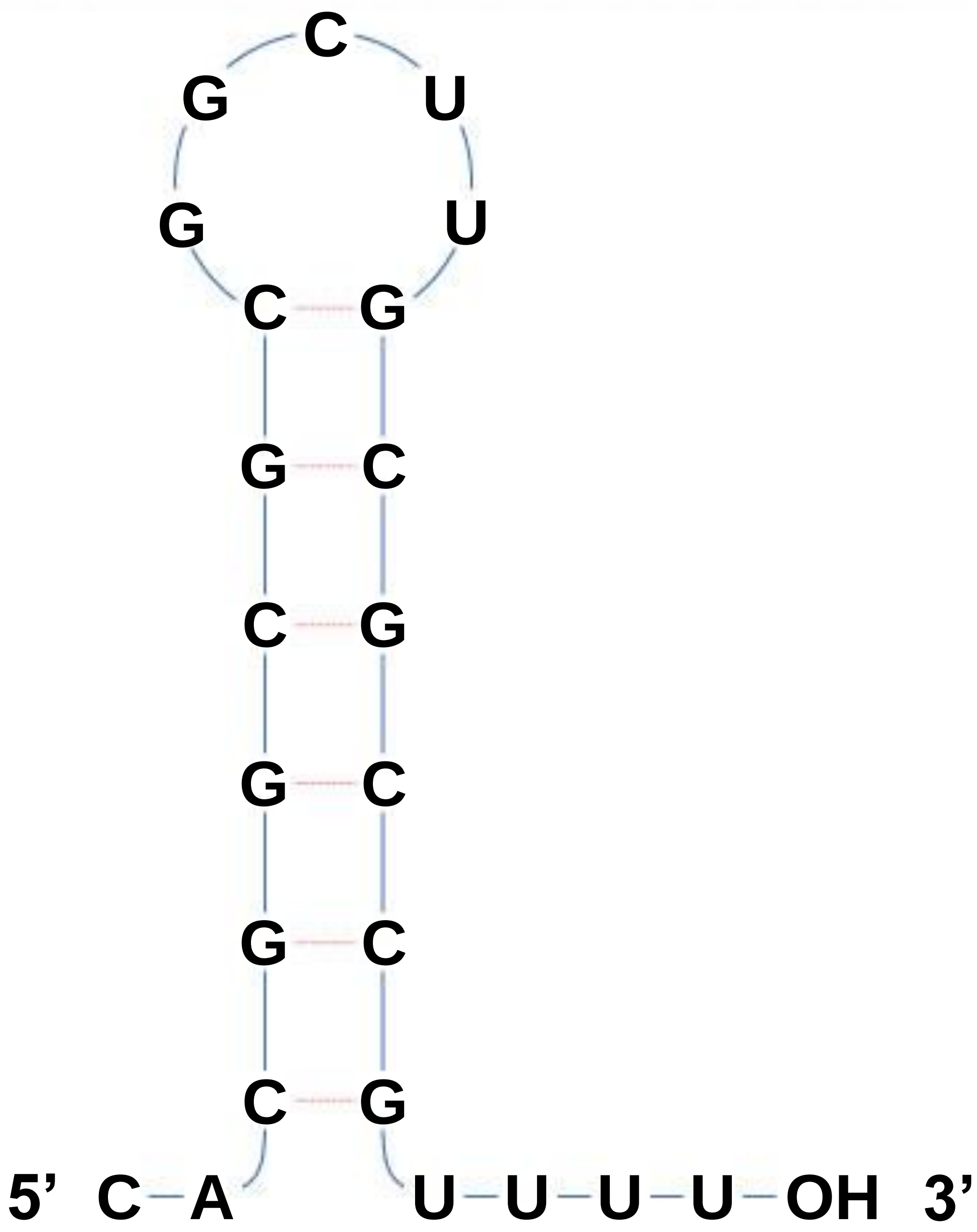
Sequence in prokaryotes cont’d

Elongation and termination phase

- Elongation: RNA polymerase adds nucleotides 5′ to 3′ at a rate of ~ 50 nucleotides/second beginning at start site. Active portion = transcription bubble.
- Completed mRNA strand exits bubble as it is finished.
- Termination: RNA polymerase stops when the RNA produces a hairpin loop.



Non-template strand
Template strand
DNA
5’
3’
3’
5’
3’
Rewinding
Unwinding
mRNA
5'
RNA polymerase
RNA-DNA hybrid helix


C
G
U
G
U
C
G
G
C
C
G
G
C
G
C
C
G
5’ C—A
U—U—U—U—OH 3’

Transcription

- Sequence in eukaryotes is basically the same; 3 differences:
  - 3 types of RNA polymerase – RNA pol I produces rRNA, RNA pol II produces mRNA, and RNA pol III produces tRNA.
  - Promoters are more complex and include sites for basal transcription factors. These replace the Sigma protein in prokaryotes, i.e. bind to DNA and open the helix. This allows for more control.

Transcription

- Sequence in eukaryotes – differences cont'd
  - Posttranscriptional modifications
    - Addition of a 5' cap (adenine or guanine + methyl-GTP and a "poly A tail", fig 16.7.

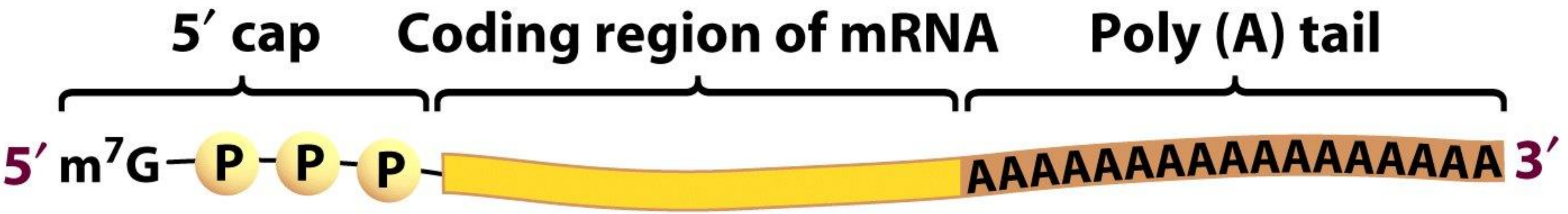


Figure 16-7 Biological Science, 2/e

Translation

Prokaryotes

Initiation phase

A small sequence of rRNA on the ribosome binds to a complementary sequence on the mRNA with the help of intiation factors.

*The start codon, AUG is exposed.

tRNA with a sequence that is complementary to the codon (=anticodon) attaches to the codon and releases its amino acid. **Each tRNA can carry only 1 type of amino acid.

Early model of tRNA function

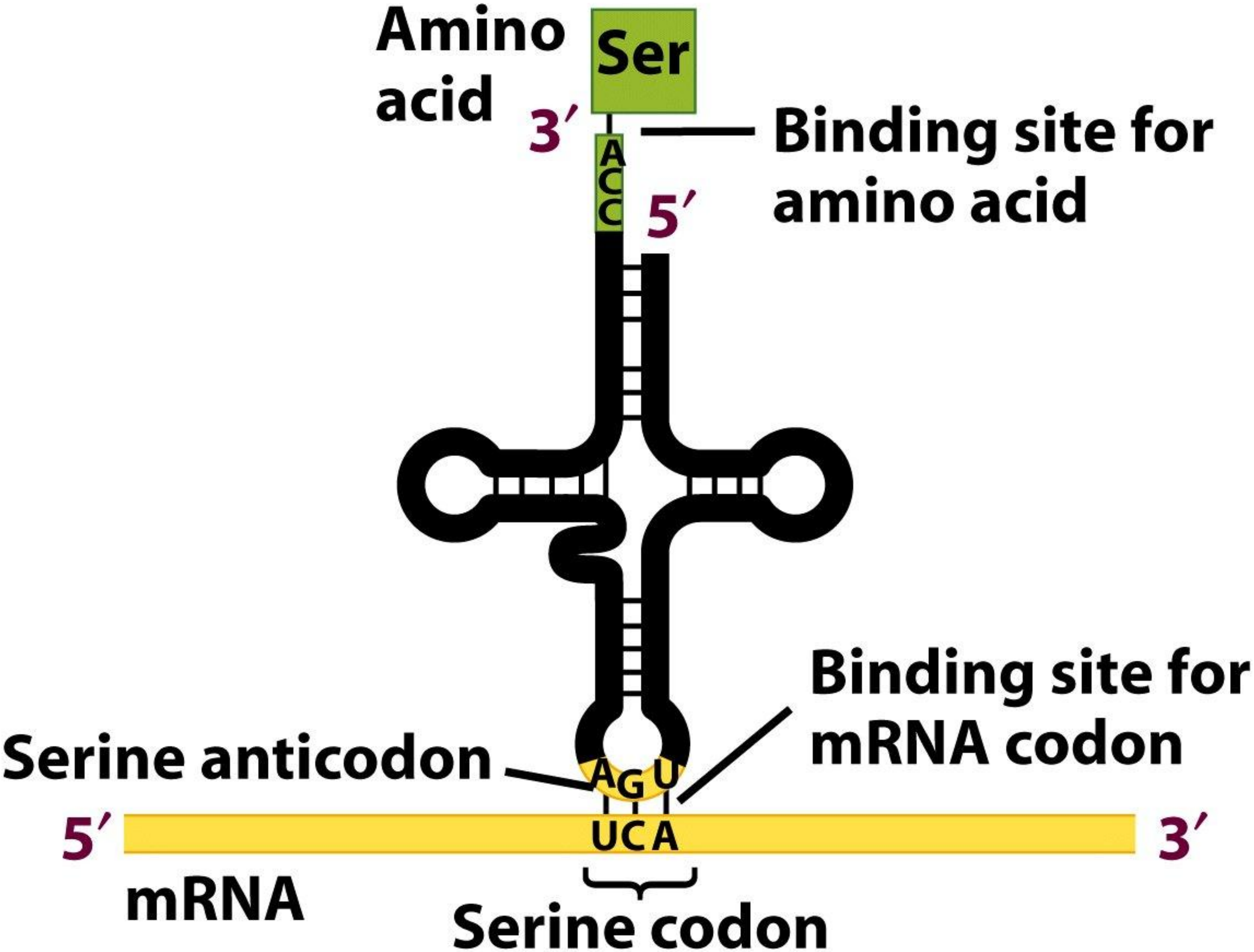


Figure 16-12beps Biological Science, 2/e

Translation

Prokaryotes

Elongation and termination phase

- Ribosome moves down 1 codon at a time and specific tRNA's bring their amino acids to the chain.
- Amino acids are joined by <u>peptide bonds</u> to form the protein.
- 3 sites on the ribosome (APE): A = tRNA binding site, P = site of peptide bond formation, E = exit site for empty tRNA's.

Diagram of ribosome during translation

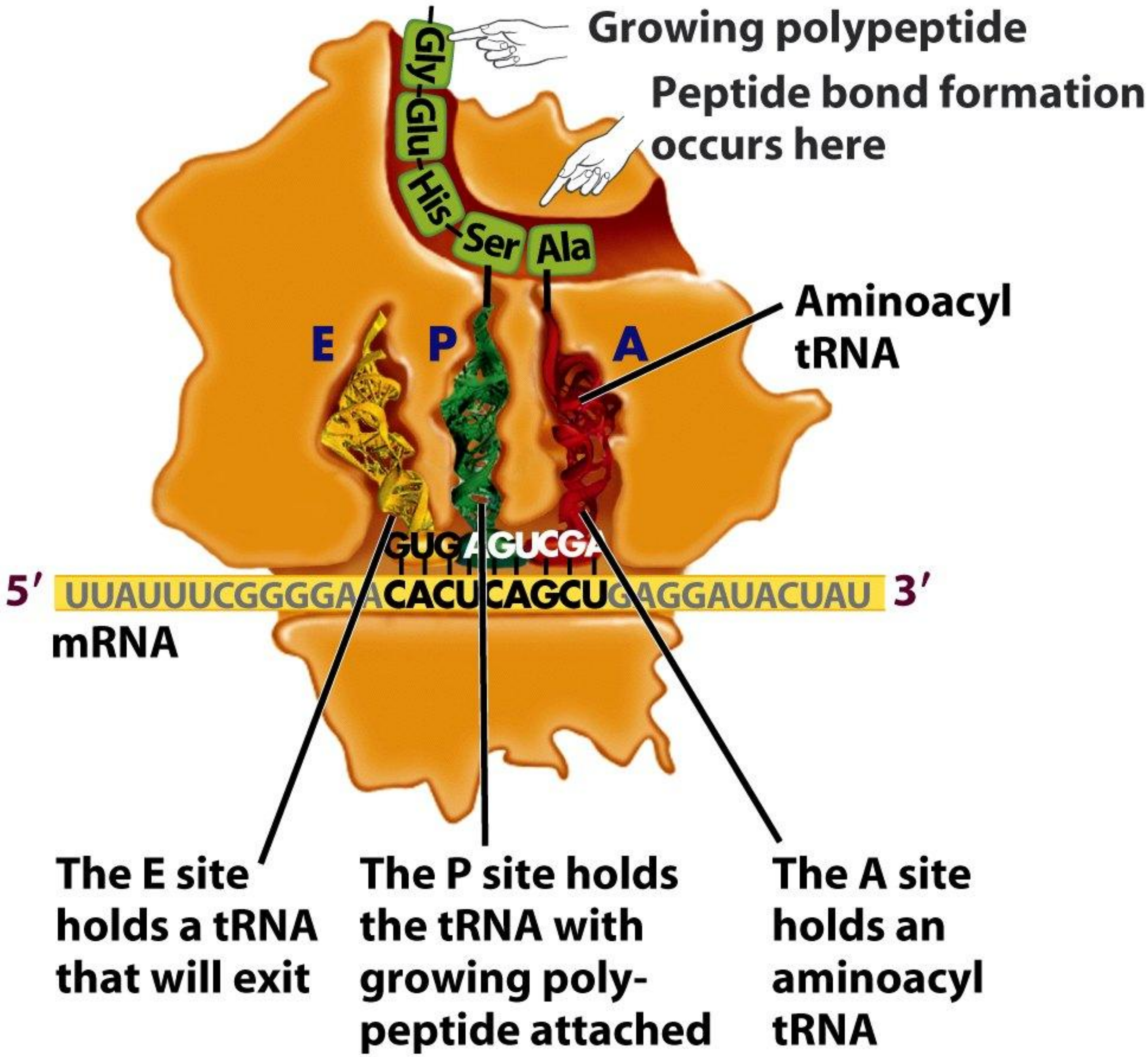


Figure 16-13b Biological Science, 2/e

Translation

- Prokaryotes cont'd
  - Elongation and termination phase
    - Translation is terminated when the ribosome reaches a stop codon.

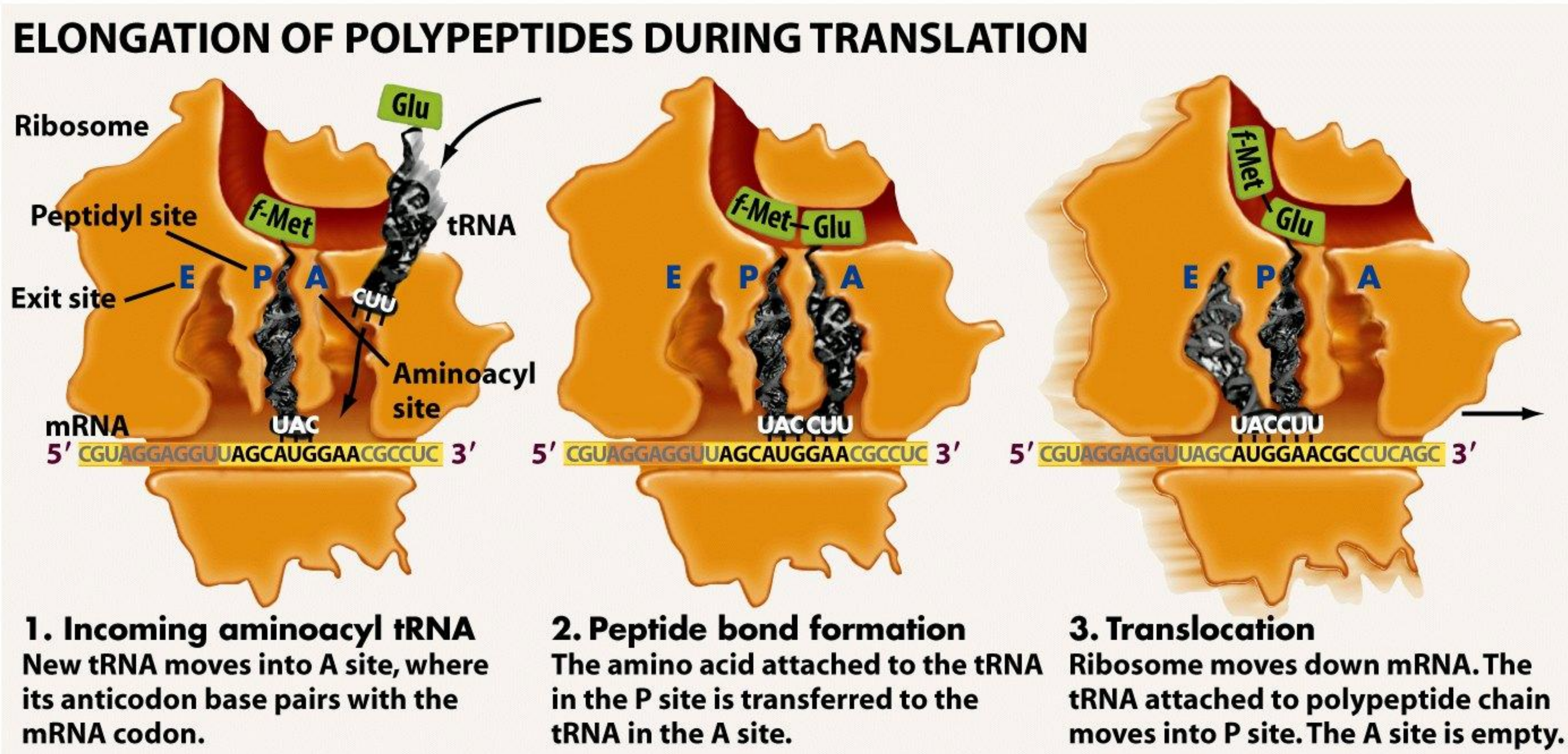


Figure 16-15 part 1 Biological Science, 2/e

4. Incoming aminoacyl tRNA
New tRNA moves into A site, where its anticodon base pairs with the mRNA codon.

5. Peptide bond formation
The polypeptide chain attached to the tRNA in the P site is transferred to the tRNA in the A site.

6. Translocation
Ribosome moves down mRNA. The tRNA attached to polypeptide chain moves into P site. Empty tRNA from P site moves to E site, where tRNA is ejected. The A site is empty again.

Figure 16-15 part 2 Biological Science, 2/e

Translation

Eukaryotes

- Eukaryotic genes contain sequences that do not contain codons = INTRONS; sequences that contain codons are EXONS.
- mRNA sequences contain the same introns and exons.

Noncoding regions must be removed from RNA transcripts.

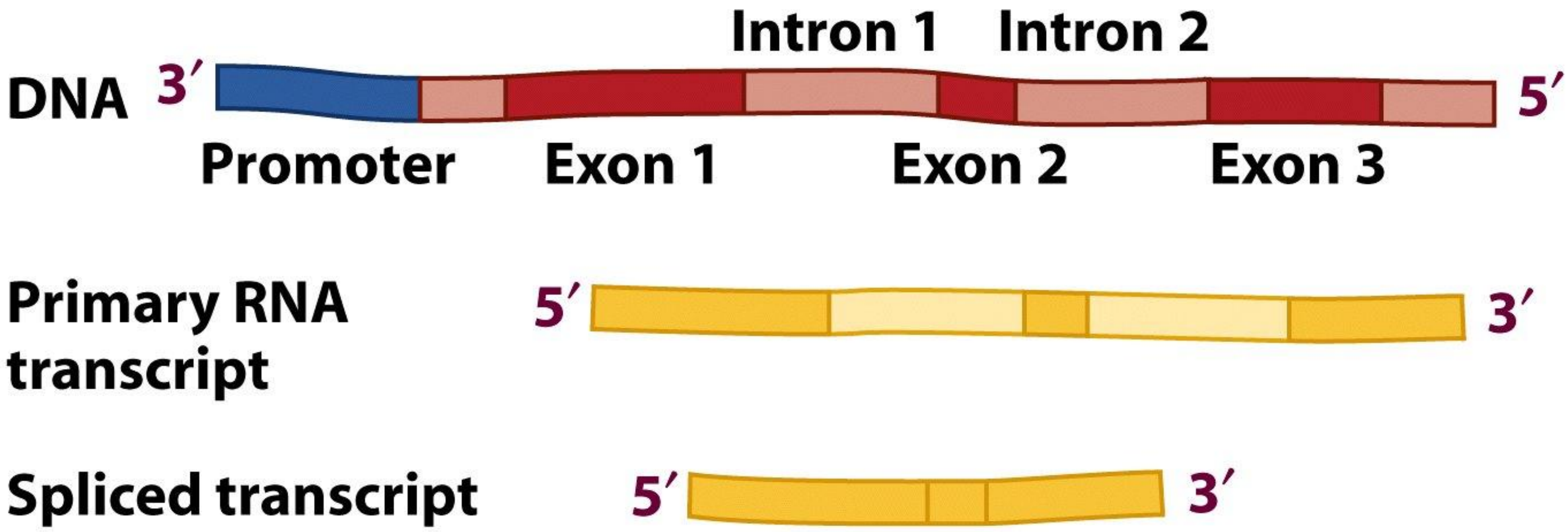


Figure 16-6a Biological Science, 2/e

Translation

Eukaryotes cont'd

Introns are removed after mRNA synthesis and exons are joined together = RNA splicing.

Why? Alternate splicing can produce different proteins from the same gene sequence. 30,000 genes can be used to produce 120,000 mRNA's.

Noncoding regions must be removed from RNA transcripts.

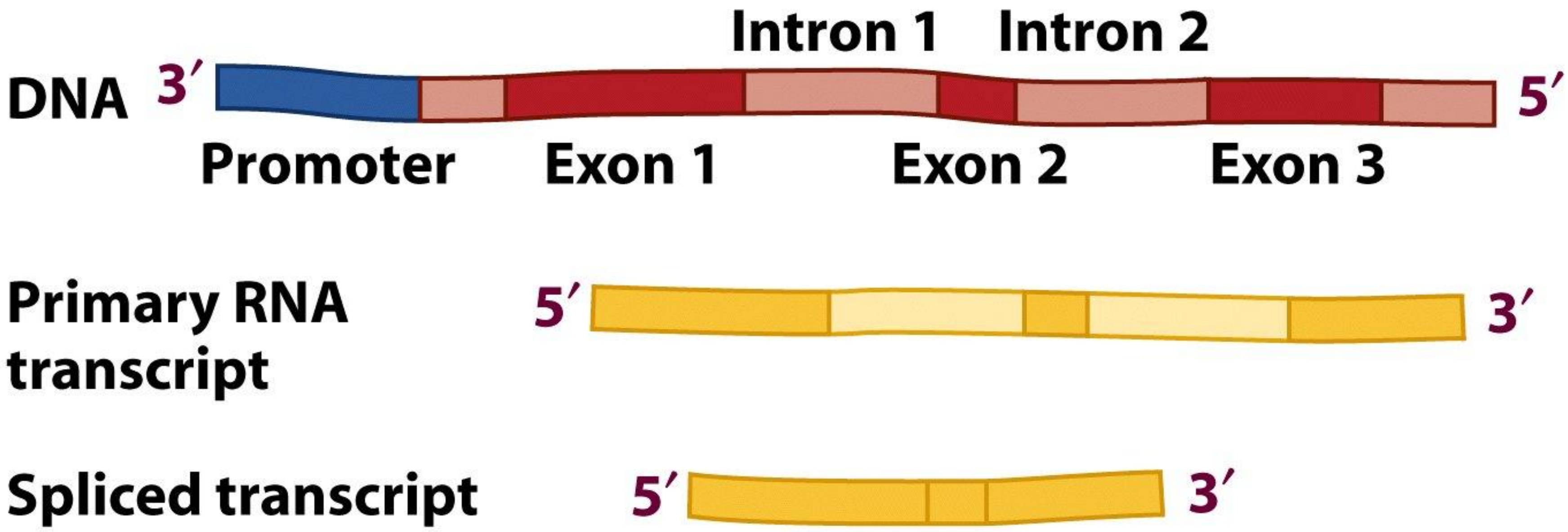


Figure 16-6a Biological Science, 2/e

Mutations

Point mutation

A change in a single nucleotide can result from errors in DNA replication or from exposure to mutagenic toxins.

In some cases, an abnormal protein is made that causes a disease process.

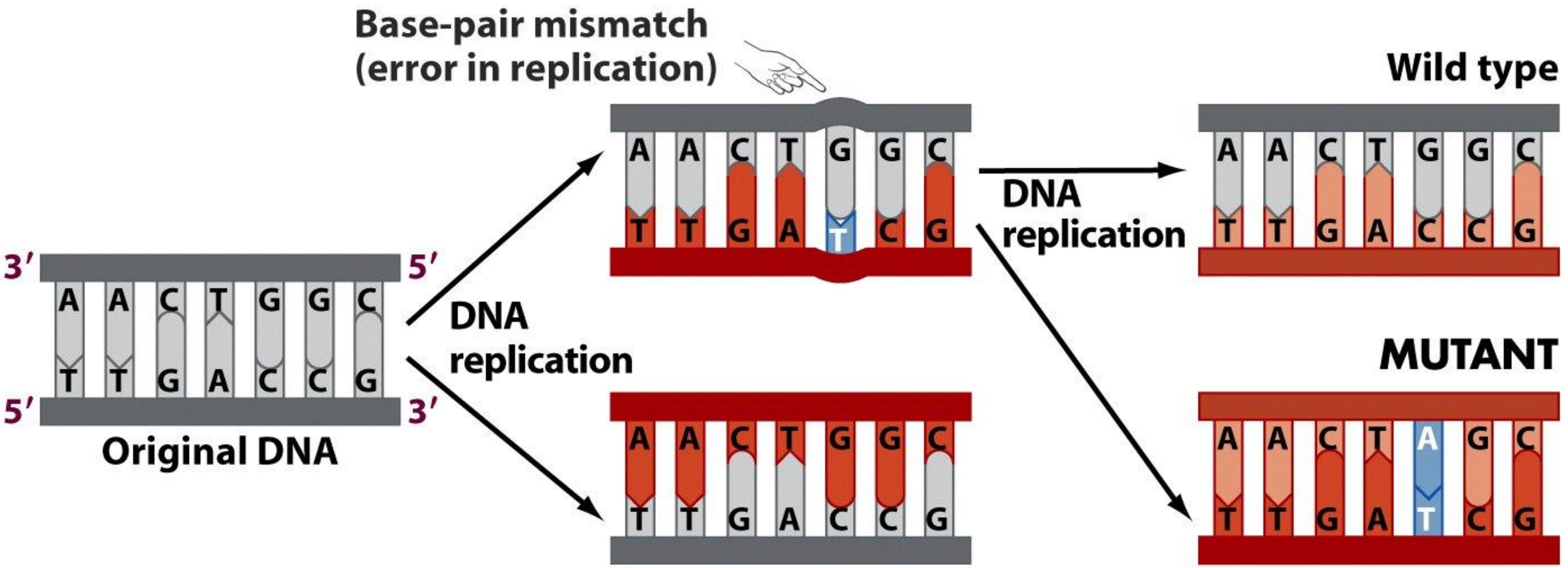


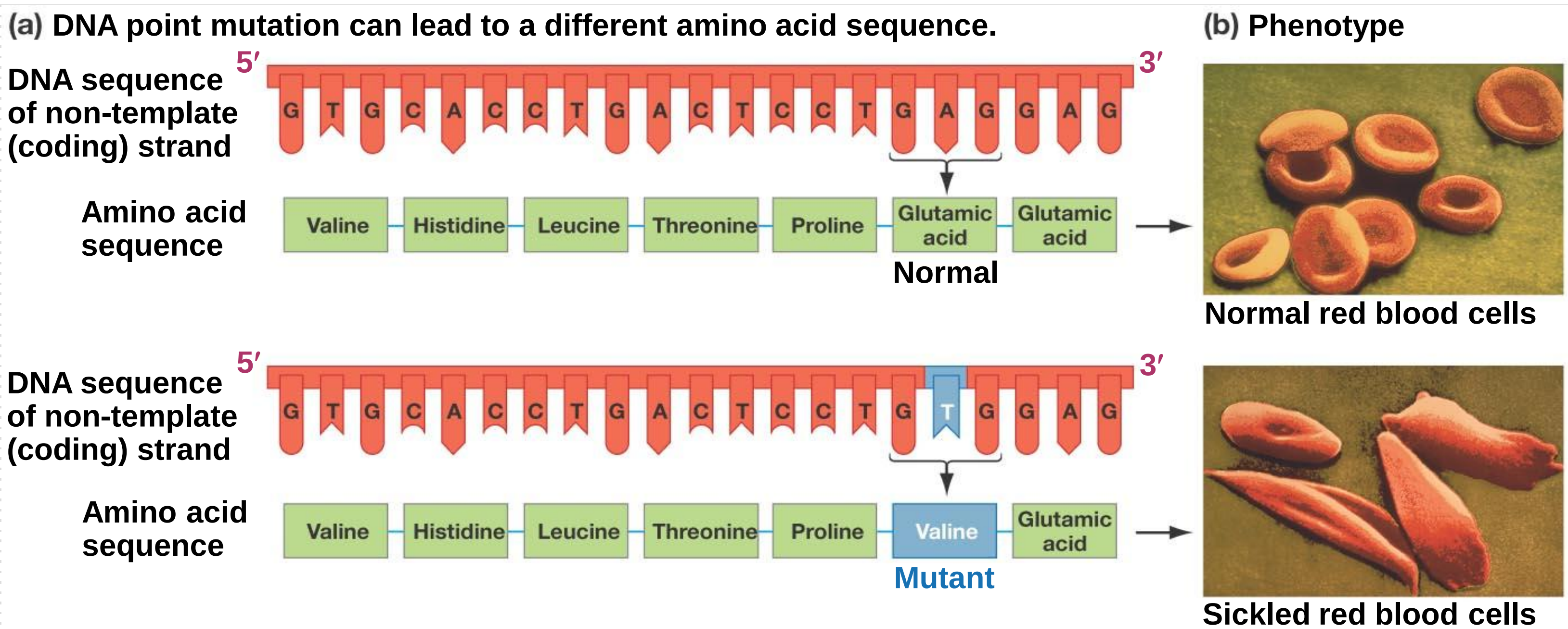
Figure 16-18 Biological Science, 2/e

Point Mutations

A single base change is called a **point mutation**.

Point mutations can result from errors in DNA replication.

Sickle-Cell Disease Results from a Point Mutation in the Gene for Hemoglobin



Point Mutations

A point mutation that causes a change in the amino acid sequence of the protein is called a **missense mutation**. Such mutations are often deleterious, meaning they reduce an individual's fitness.

For example, sickle-cell disease results from a missense mutation in the hemoglobin gene.

Mutations that do not change the amino acid sequence of the protein are known as **silent mutations**. Such mutations are said to be neutral, as they do not affect an individual's fitness.

Types of point mutations

	Definition	Example	Consequence
		Original DNA sequence — TAT TGG CTA GTA CAT Original polypeptide — Tyr–Trp–Leu–Val–His	
Silent	Change in nucleotide that does not change amino acid specified by codon	TAC TGG CTA GTA CAT Tyr–Trp–Leu–Val–His	Change in genotype but no change in phenotype
Missense (Replacement)	Change in nucleotide that changes amino acid specified by codon	TAT TGT CTA GTA CAT Tyr–Cys–Leu–Val–His	Change in primary structure of protein
Nonsense	Change in nucleotide that results in early stop codon	TAT TGA CTA GTA CAT Tyr STOP	Premature termination—polypeptide is truncated
Frameshift	Addition or deletion of a nucleotide	TAT TCG GCT AGT ACA T Tyr–Ser–Ala–Ser–Thr	Reading frame is shifted—massive missense

Figure 16-20a Biological Science, 2/e

Mutations

<u>Gene duplication</u> – extra copy of gene is added to one chromosome during cross over.

<u>Gene inversion</u> – piece of DNA is flipped over during cross over.

<u>Gene deletion</u> – loss of DNA.

Other types of mutations

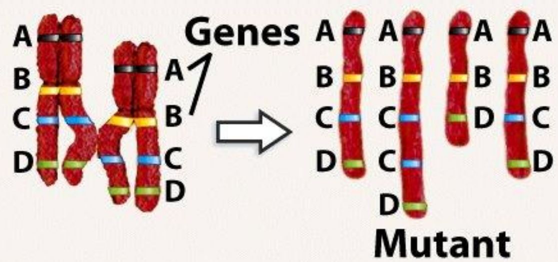
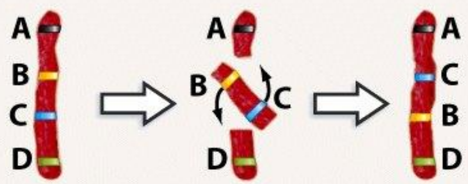
	Definition	Example	Consequence
Gene duplication	Addition of a small chromosome segment due to an error during crossing over at meiosis I—homologs do not align correctly		Produces an extra copy or deletion of one or more genes. Families of related genes arise by gene duplication.
Chromosome inversion	Change in a chromosome segment when DNA breaks in two places, flips, and rejoins		Changes gene order along chromosome. Other types of chromosome breaks can lead to deletion or addition of chromosome segments.

Figure 16-20b Biological Science, 2/e

Differences between Prokaryotic and Eukaryotic Gene Expression

- Eukaryotic genes contain introns; prokaryotic genes do not.
- Eukaryotic mRNA’s code for 1 gene; prokaryotic mRNA’s code for several related genes at one time.
- Eukaryotic mRNA must be moved to cytoplasm; prokaryotic mRNA is already in the cytoplasm and translation starts before transcription is complete.

Differences between Prokaryotic and Eukaryotic Gene Expression

Eukaryotic mRNA’s are modified for transport – RNA splicing, poly A tail; prokaryotic mRNA’s are not modified.

Eukaryote ribosomes are larger than prokaryote ribosomes and many ribosomes may participate in translation of 1 eukaryotic protein.