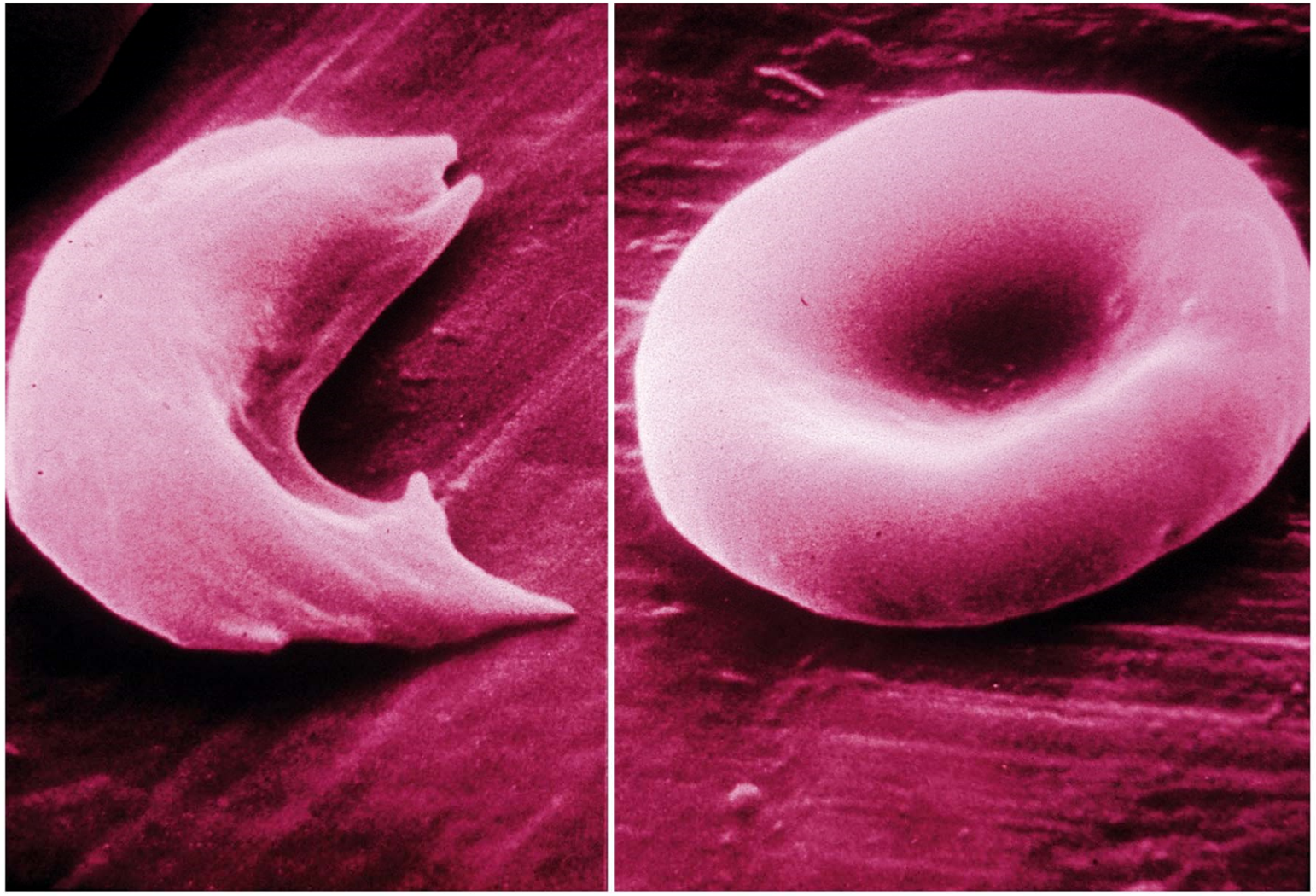
Chapter 5: Protein Synthesis

Function of protein

Structure of protein

Translation

Devastating effect of a single base pair change…



Chapter 12 Opener Principles of Genetics, 4/e

Scanning electron micrograph of sickle-shape (left) and normal red blood cells (right)

Protein

- Most genes exert their effect(s) on the phenotype of an organism through proteins, which are large macromoelcules composed of polypeptide
- Each polypeptide is a chainlike polymer assembled from different amino acids
- The amino acid sequence of each polypeptide is specified by the nucleotide sequence of a gene
- The vast functional diversity of proteins results in part from their complex three-dimensional structures

Function of Protein

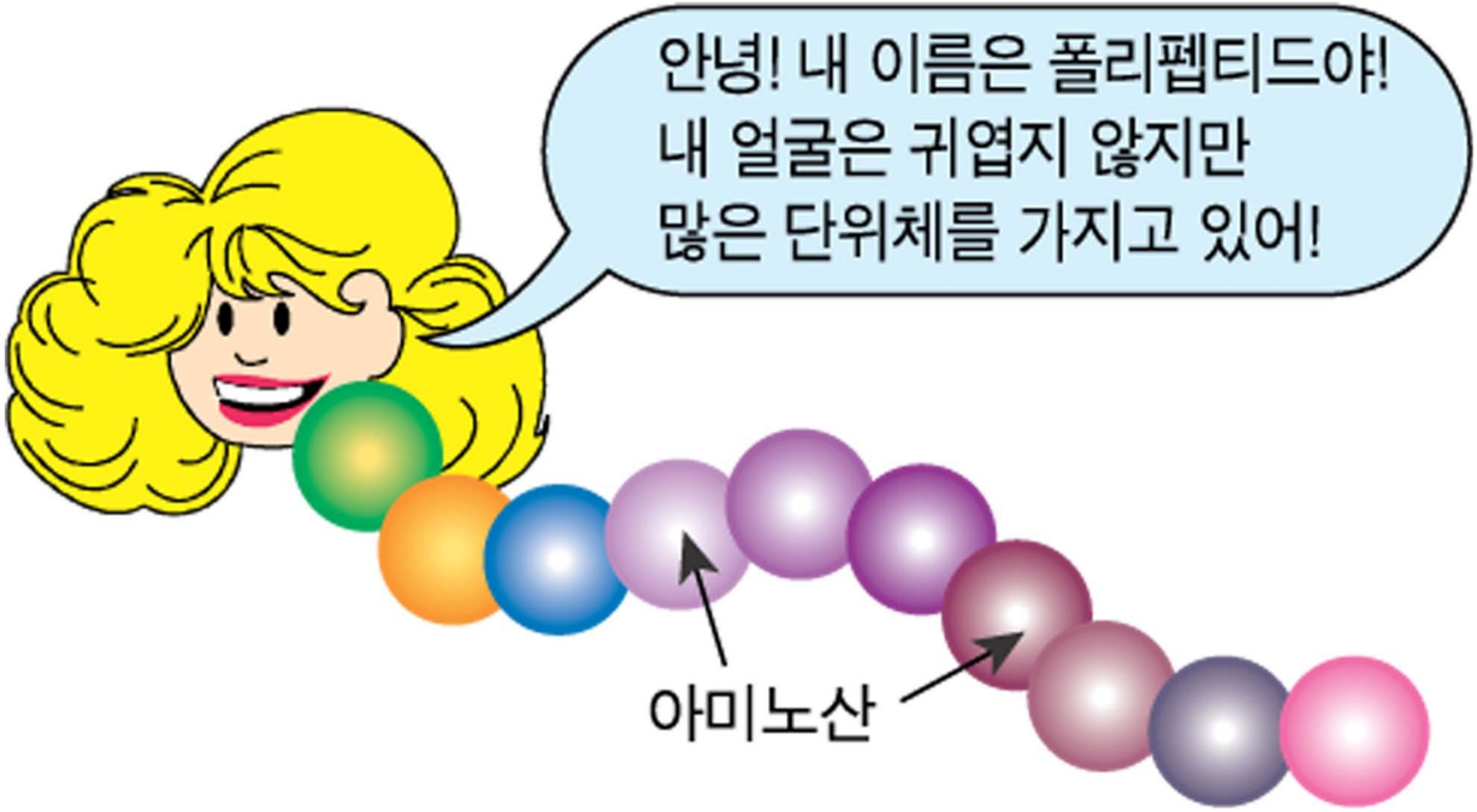
- **Role of proteins in the cell**

  1) **Structural protein**
     - forms part of a cellular structure
  2) **Enzyme**
     - carries out a chemical reaction
  3) **Regulatory protein**
     - controls the expression of a gene or the activity of other proteins
  4) **Transport protein**
     - carries other molecules across membranes or around body
     - found mostly in biological membranes (membrane transport protein)
     - hemoglaobin

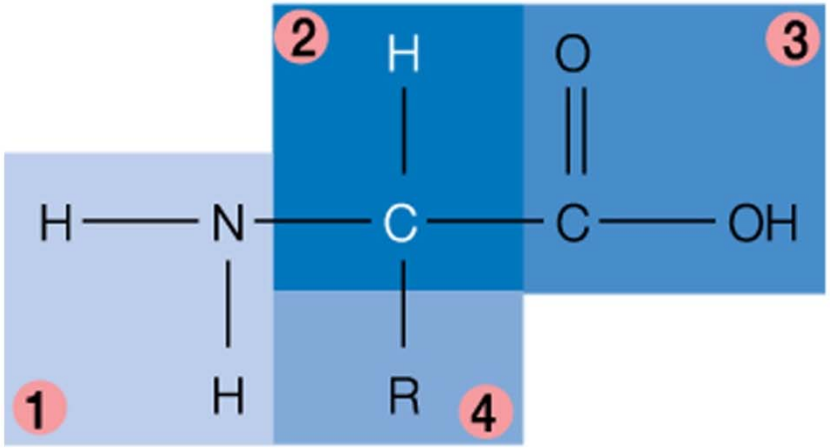
Structure of Protein

- Proteins are made by using 20 different amino acids
- Proteins are made from a **linear chain of amino acids** (=polypeptide chain)
- Amino acid has four common features

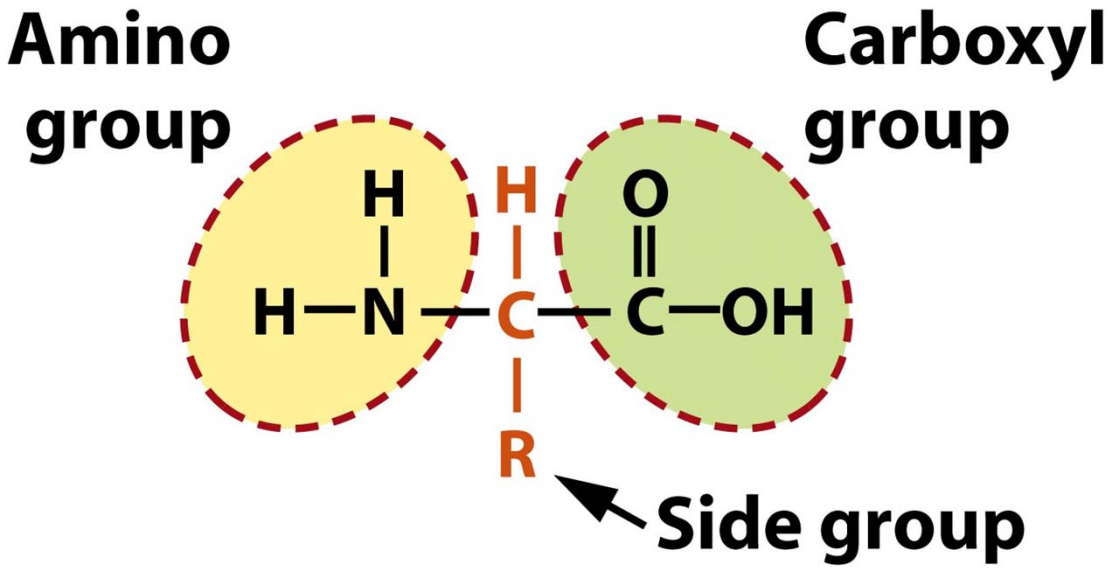
    1) NH2 (amino) group
    2) central hydrocarbon group
    3) COOH (carboxy) group
    4) R or variable group


안녕! 내 이름은 폴리펩티드야!
내 얼굴은 귀엽지 않지만
많은 단위체를 가지고 있어!
아미노산

아미노산은 공통된 특징을 가지고 있다.



1. NH_2(아미노)기
2. 중앙 탄화수소기
3. COOH(카르복시)기
4. R기(R-group) 또는 가변기



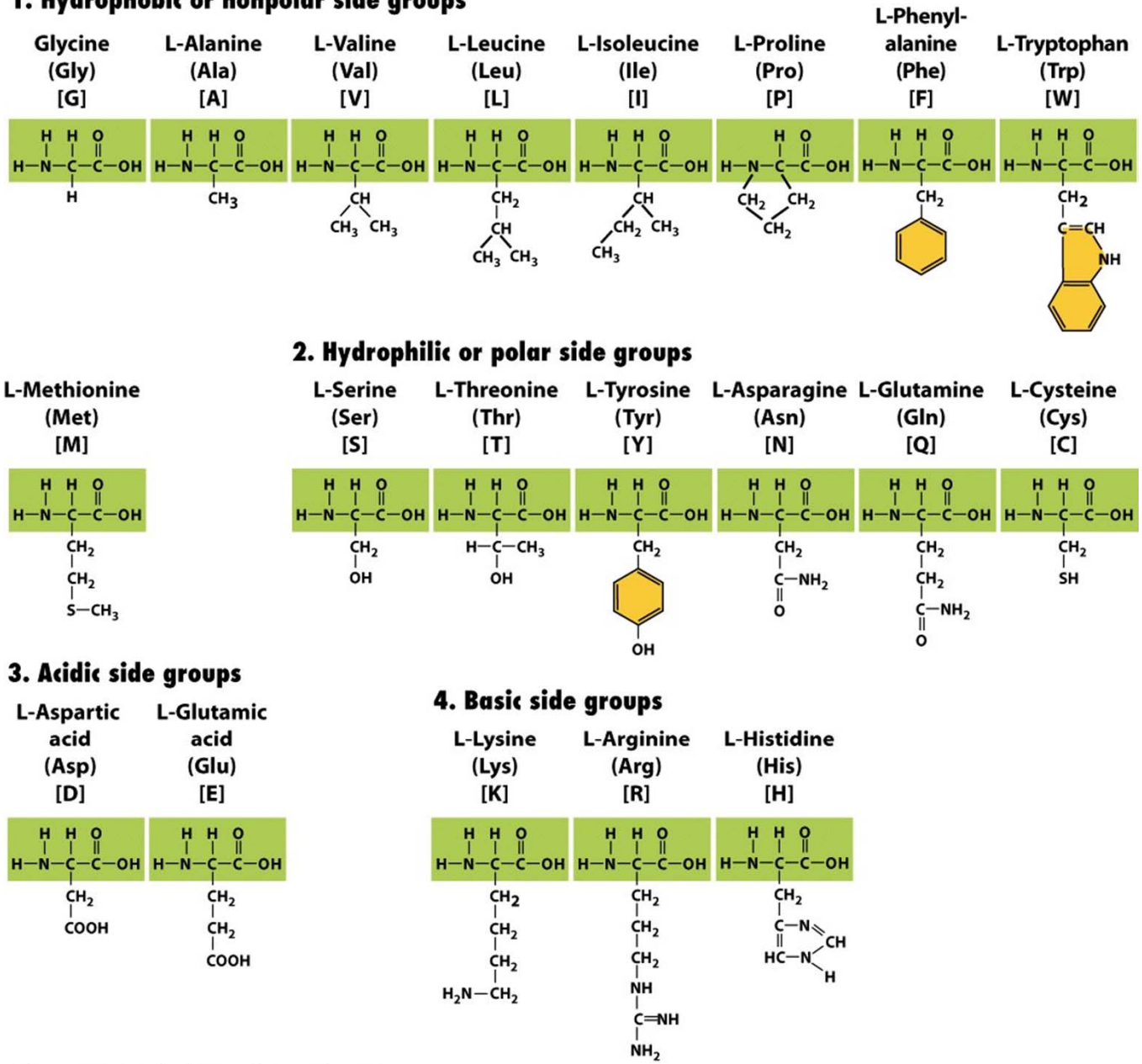


Figure 12-1 Principles of Genetics, 4/e

Structure of Protein

- Amino acids are joined together by polypeptide bonds to give a linear polymer called a polypeptide chain (N-terminus ----------C-terminus)

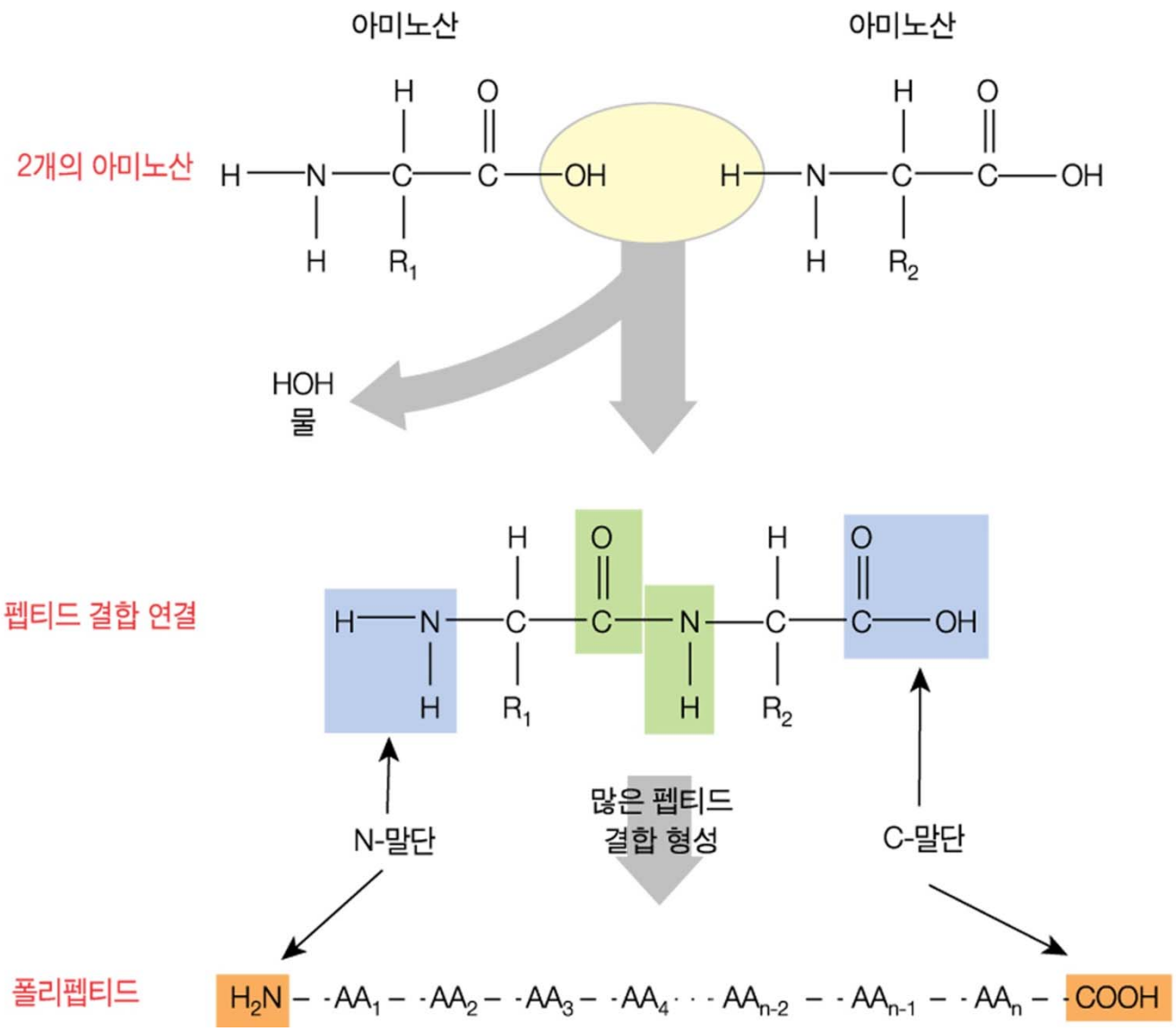
- The structures of protein

  1) Primary structure; the linear order of amino acids are arranged

  2) Secondary structure; initial folding of the primer structure by H-bonding

  3) Tertiary structure; final 3-D folding of the secondary structure by R-groups

  4) Quaternary structure; aggregation of more than one polymer of tertiary structure in final structure



Formation of a **peptide bond** between two amino acids by the removal of water. Each peptide bond connects the **amino group** of one amino acid and the **carboxyl group** of the adjacent amino acid

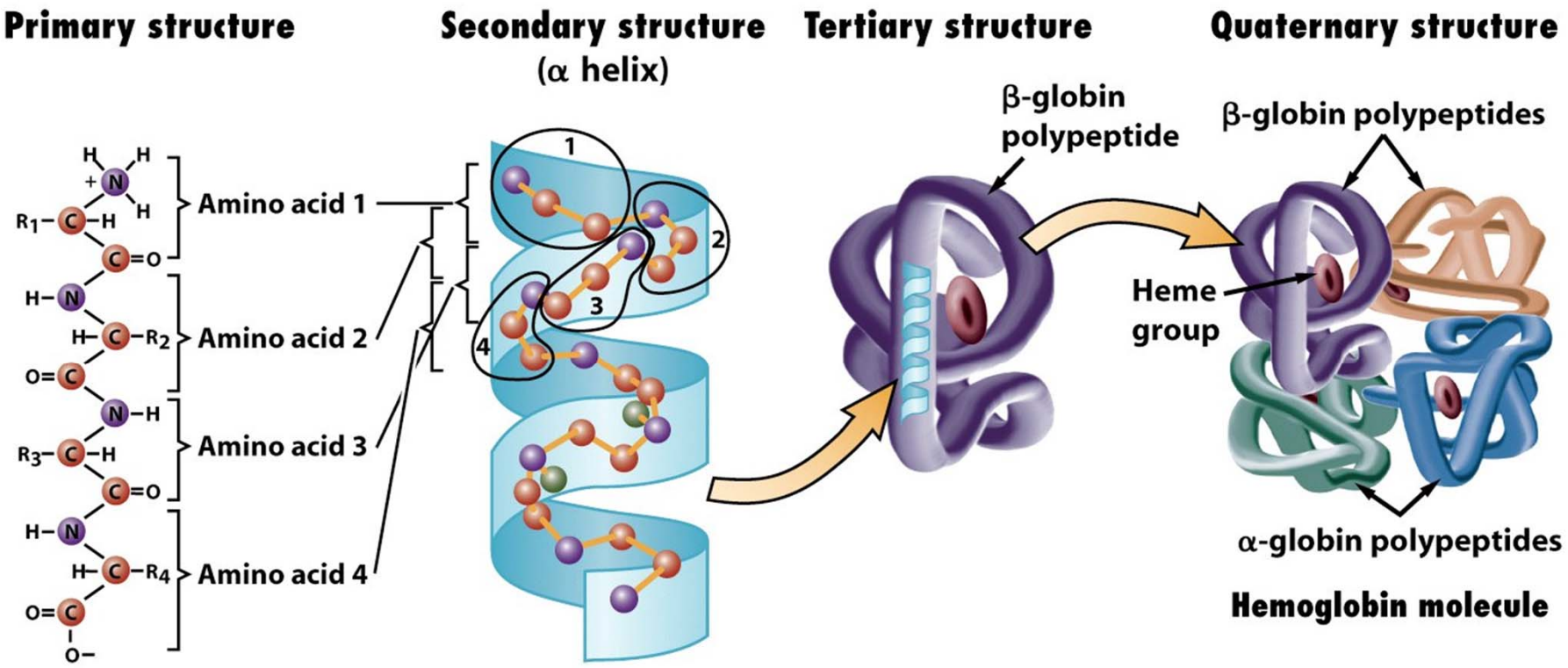


Figure 12-3 Principles of Genetics, 4/e

The four levels of organization in protein – hemoglobin as an example

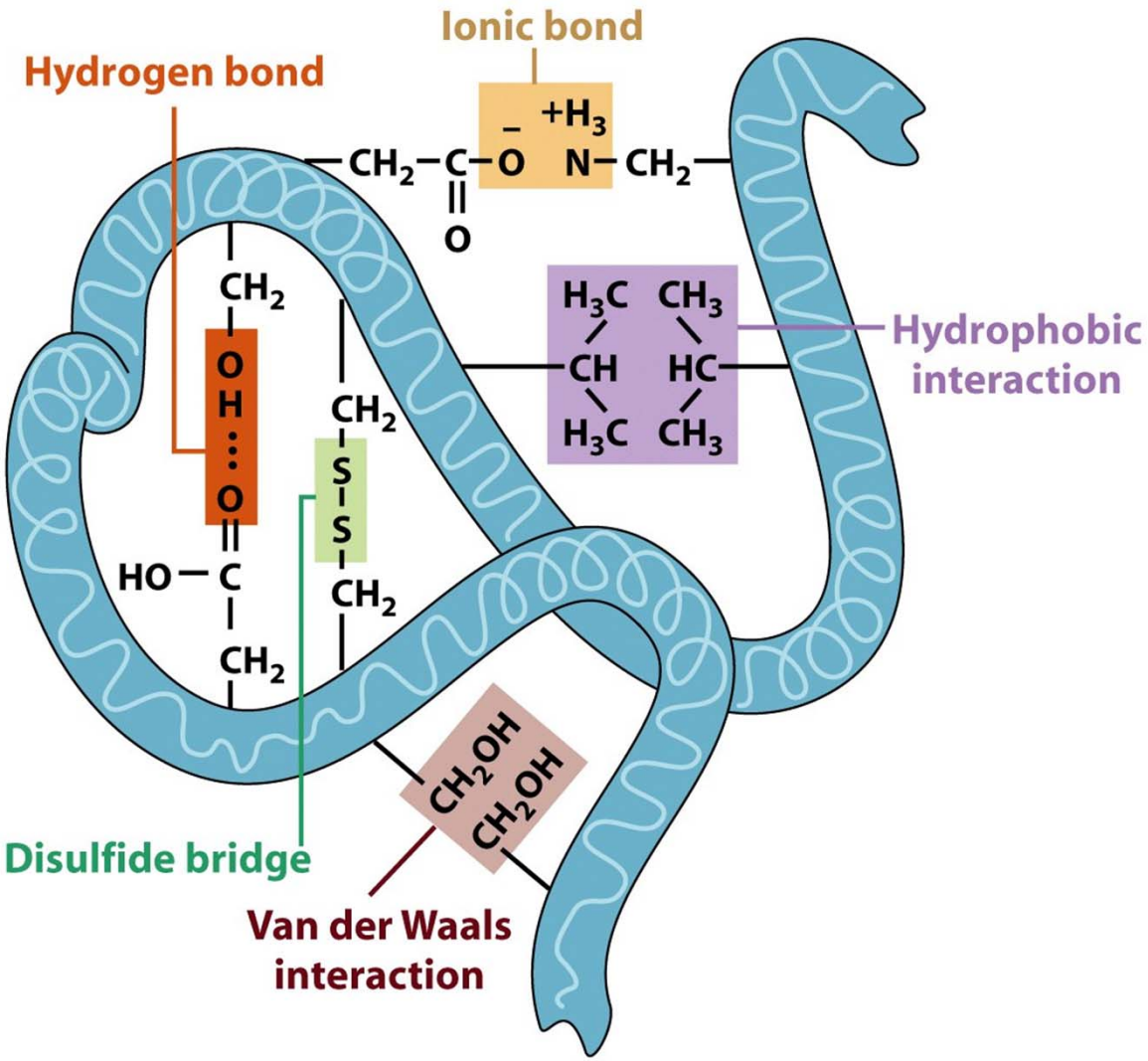
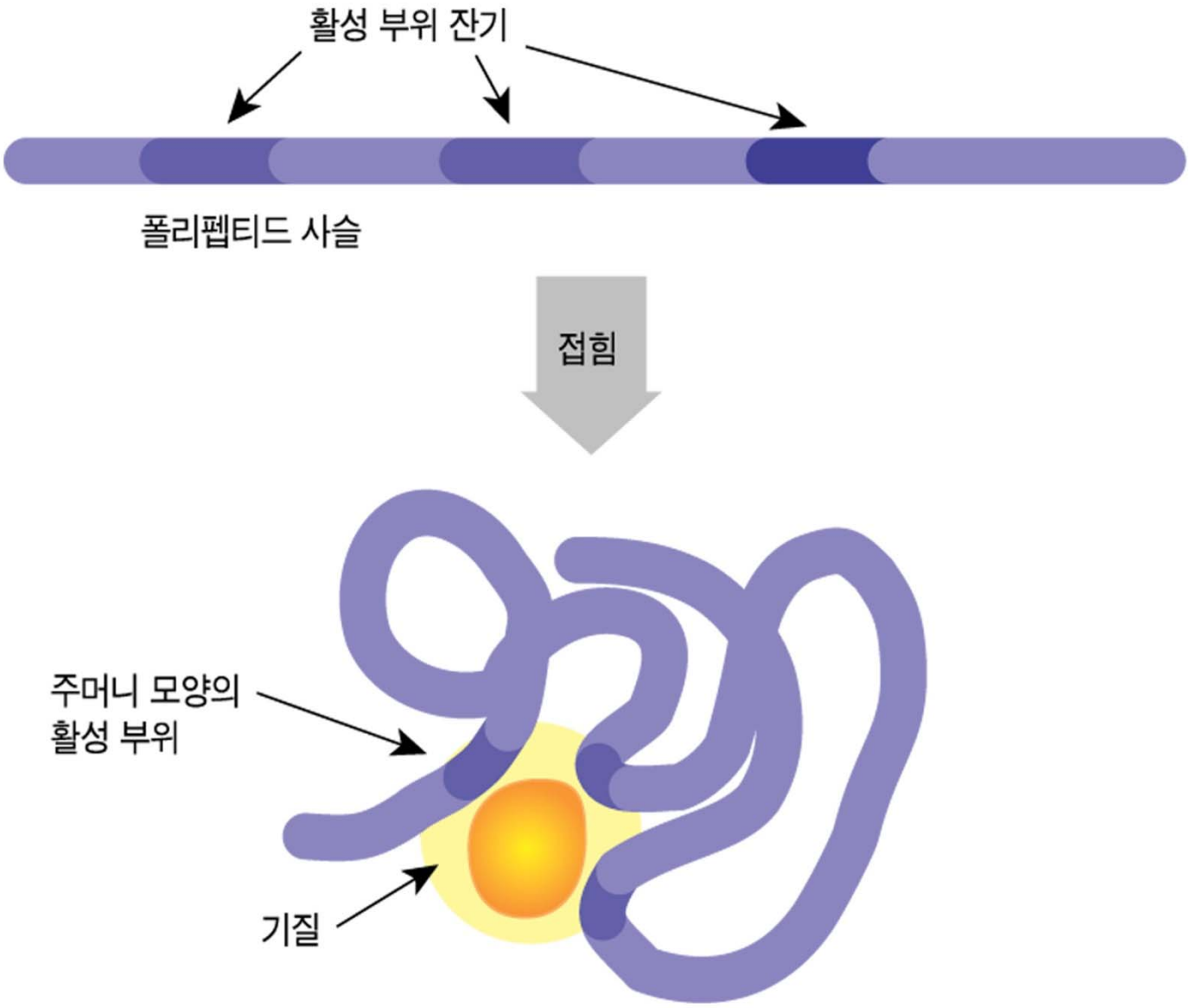


Figure 12-5 Principles of Genetics, 4/e

The four levels of organization in protein – hemoglobin as an example


활성 부위 잔기
폴리펩티드 사슬
접힘
주머니 모양의
활성 부위
기질

Overview of Protein Synthesis (Translation)

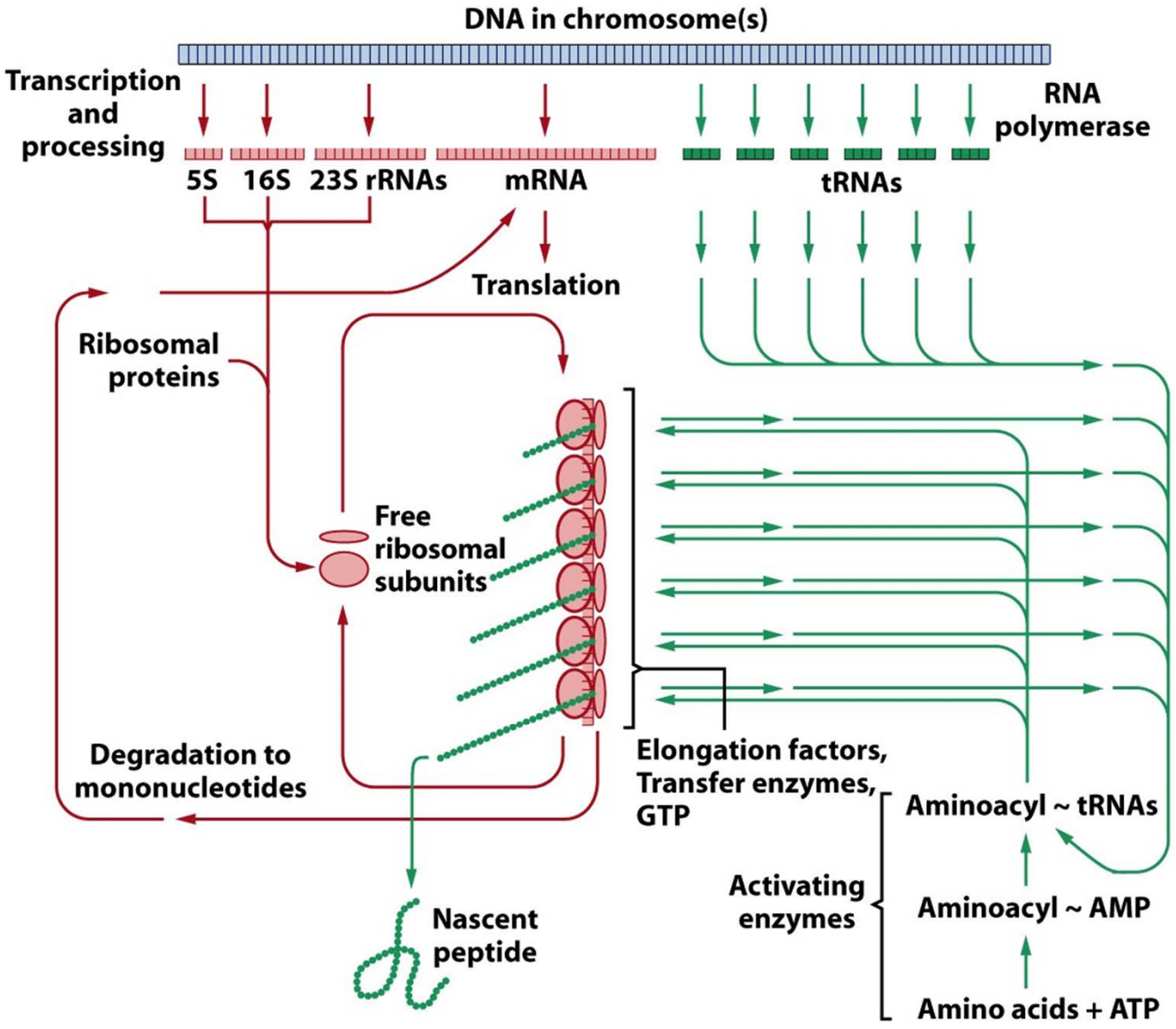


Figure 12-6 Principles of Genetics, 4/e

Major Components of the Protein Synthesis System

- Triplet codon (in messenger RNA)
- Ribosome (with rRNA)
- Transfer RNA (tRNAs)
- Amino acid (AA)

Properties of genetic code

1) Each genetic code is composed of nucleotide triplets
- Three nucleotides in mRNA specify one amino acid in the polypeptide products

2) The genetic code is non overlapping
- Each nucleotide in mRNA belongs to just one codon

3) The genetic code is degenerate (degeneracy)
- All but two of the amino acids are specified by more than one codon

4) The genetic code contains start and stop codons
- Specific codons are used to initiate and to terminate polypeptide chains

5) The genetic code is nearly universal
- With minor exceptions, the codons have the same meaning in all living organisms, from viruses to human

Triplet Codons

Group of three RNA or DNA bases which encodes a single AA

두 번째 (중간) 염기

첫 번째 염기	U	C	A	G	세 번째 염기
U	UUU Phe UUC Phe UUA Leu UUG Leu	UCU Ser UCC Ser UCA Ser UCG Ser	UAU Tyr UAC Tyr UAA STOP UAG STOP	UGU Cys UGC Cys UGA STOP UGG Trp	U C A G
C	CUU Leu CUC Leu CUA Leu CUG Leu	CCU Pro CCC Pro CCA Pro CCG Pro	CAU His CAC His CAA Gln CAG Gln	CGU Arg CGC Arg CGA Arg CGG Arg	U C A G
A	AUU Ile AUC Ile AUA Ile AUG Met	ACU Thr ACC Thr ACA Thr ACG Thr	AAU Asn AAC Asn AAA Lys AAG Lys	AGU Ser AGC Ser AGA Arg AGG Arg	U C A G
G	GUU Val GUC Val GUA Val GUG Val	GCU Ala GCC Ala GCA Ala GCG Ala	GAU Asp GAC Asp GAA Glu GAG Glu	GGU Gly GGC Gly GGA Gly GGG Gly	U C A G

Cracking the genetic code

1968, Nirenberg and Khorana: found out meaning of all 64 triplet codons

TABLE 12.1

The Genetic Code[a]

First (5′) letter	Second letter: U	Second letter: C	Second letter: A	Second letter: G	Third (3′) letter
U	UUU, UUC: Phe	UCU, UCC, UCA, UCG: Ser	UAU, UAC: Tyr	UGU, UGC: Cys	U
	UUA, UUG: Leu		UAA: *Ochre* (terminator); UAG: *Amber* (terminator)	UGA: *Opal* (terminator); UGG: Trp	C
					A
					G
C	CUU, CUC, CUA, CUG: Leu	CCU, CCC, CCA, CCG: Pro	CAU, CAC: His; CAA, CAG: Gln	CGU, CGC, CGA, CGG: Arg	U
					C
					A
					G
A	AUU, AUC, AUA: Ileu	ACU, ACC, ACA, ACG: Thr	AAU, AAC: Asn; AAA, AAG: Lys	AGU, AGC: Ser; AGA, AGG: Arg	U
	AUG: *Met* (initiator)				C
					A
					G
G	GUU, GUC, GUA, GUG: Val	GCU, GCC, GCA, GCG: Ala	GAU, GAC: Asp; GAA, GAG: Glu	GGU, GGC, GGA, GGG: Gly	U
					C
					A
					G

= Polypeptide chain initiation codon

= Polypeptide chain termination codon

[a]Each triplet nucleotide sequence or codon refers to the nucleotide sequence in **mRNA** (not DNA) that specifies the incorporation of the indicated amino acid or polypeptide chain termination.

Table 12-1 Principles of Genetics, 4/e

Degeneracy: a way of minimizing mutational lethal?

TABLE 12.1

The Genetic Code[a]

First (5′) letter	Second letter: U	Second letter: C	Second letter: A	Second letter: G	Third (3′) letter
U	UUU Phe	UCU Ser	UAU Tyr	UGU Cys	U
	UUC Phe	UCC Ser	UAC Tyr	UGC Cys	C
	UUA Leu	UCA Ser	UAA *Ochre* (terminator)	UGA *Opal* (terminator)	A
	UUG Leu	UCG Ser	UAG *Amber* (terminator)	UGG Trp	G
C	CUU Leu	CCU Pro	CAU His	CGU Arg	U
	CUC Leu	CCC Pro	CAC His	CGC Arg	C
	CUA Leu	CCA Pro	CAA Gln	CGA Arg	A
	CUG Leu	CCG Pro	CAG Gln	CGG Arg	G
A	AUU Ileu	ACU Thr	AAU Asn	AGU Ser	U
	AUC Ileu	ACC Thr	AAC Asn	AGC Ser	C
	AUA Ileu	ACA Thr	AAA Lys	AGA Arg	A
	AUG *Met* (initiator)	ACG Thr	AAG Lys	AGG Arg	G
G	GUU Val	GCU Ala	GAU Asp	GGU Gly	U
	GUC Val	GCC Ala	GAC Asp	GGC Gly	C
	GUA Val	GCA Ala	GAA Glu	GGA Gly	A
	GUG Val	GCG Ala	GAG Glu	GGG Gly	G

Complete degeneracy

Partial degeneracy

= Polypeptide chain initiation codon

= Polypeptide chain termination codon

[a]Each triplet nucleotide sequence or codon refers to the nucleotide sequence in **mRNA** (not DNA) that specifies the incorporation of the indicated amino acid or polypeptide chain termination.

Table 12-1 Principles of Genetics, 4/e

Degeneracy and recognization of codons by tRNA

Several tRNAs exist for certain amino acids, and some tRNA recognize more than one codon -> **Wobble Hypothesis** (F. Crick)

TABLE 12.2

Base-Pairing Between the 5′ Base of the Anticodons of tRNAs and the 3′ Base of Codons of mRNAs According to the Wobble Hypothesis

Base in Anticodon	Base in Codon
G	U or C
C	G
A	U
U	A or G
I	A, U, or C

Table 12-2 Principles of Genetics, 4/e

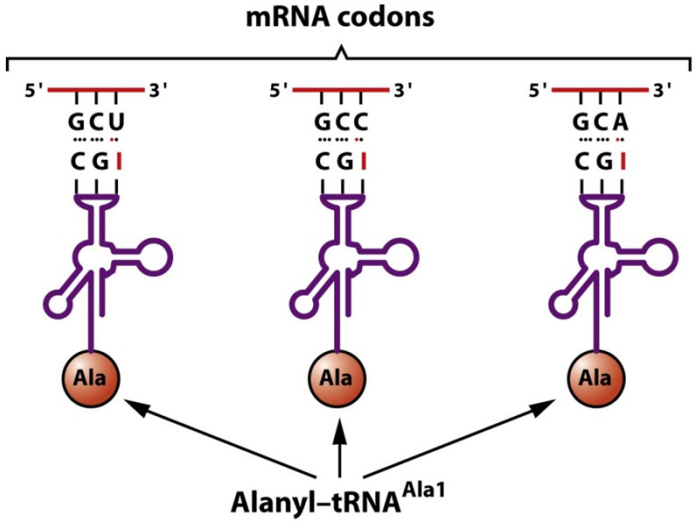


Figure 12-21 Principles of Genetics, 4/e

-Several type of base paring at the third codon base in codon-anticodon interaction

-Codon/anticodon paring of mRNA/tRNA follows wobble rules

Ribosomes

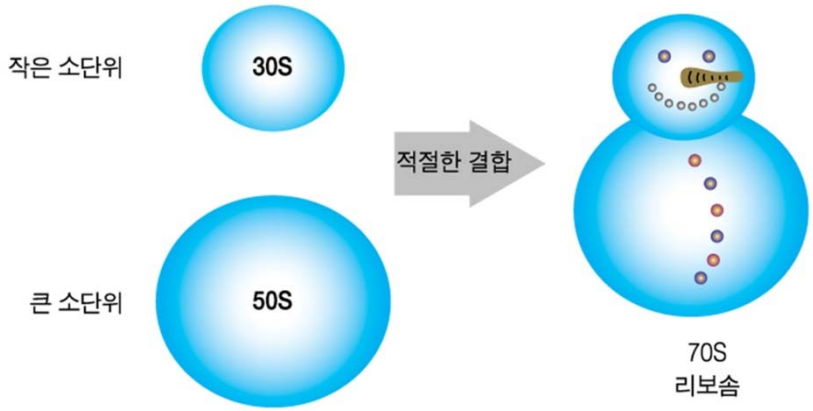
- Codons are decoded to protein by ribosome (decoding machine) that binds mRNA and translates it

- Components of ribosomes (in bacteria)

  30S small subunit (16S rRNA + 21 proteins)+50S large subunit (5S rRNA + 23S rRNA + 31 proteins) = 70S ribosome

- Ribosome binds to mRNA, moves along it, adding new amino acid to the growing polypeptide chain each time it read a codon from the massage

Macromolecule Composition of Ribosome in Bacteria



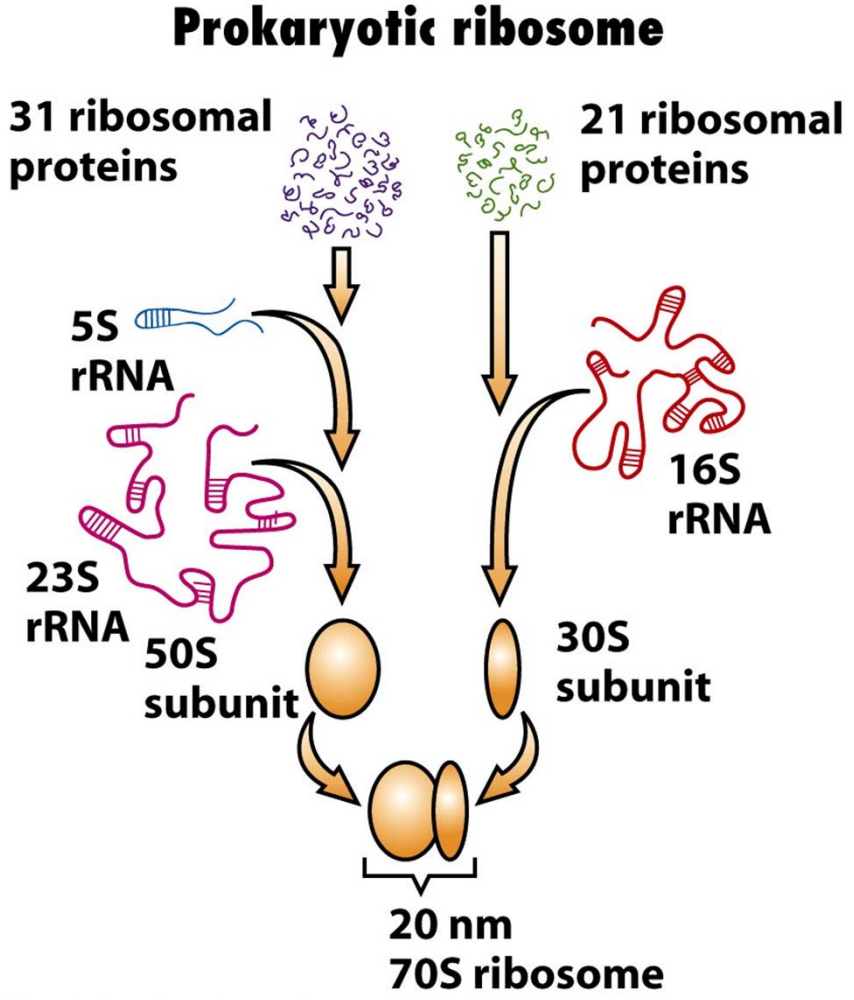
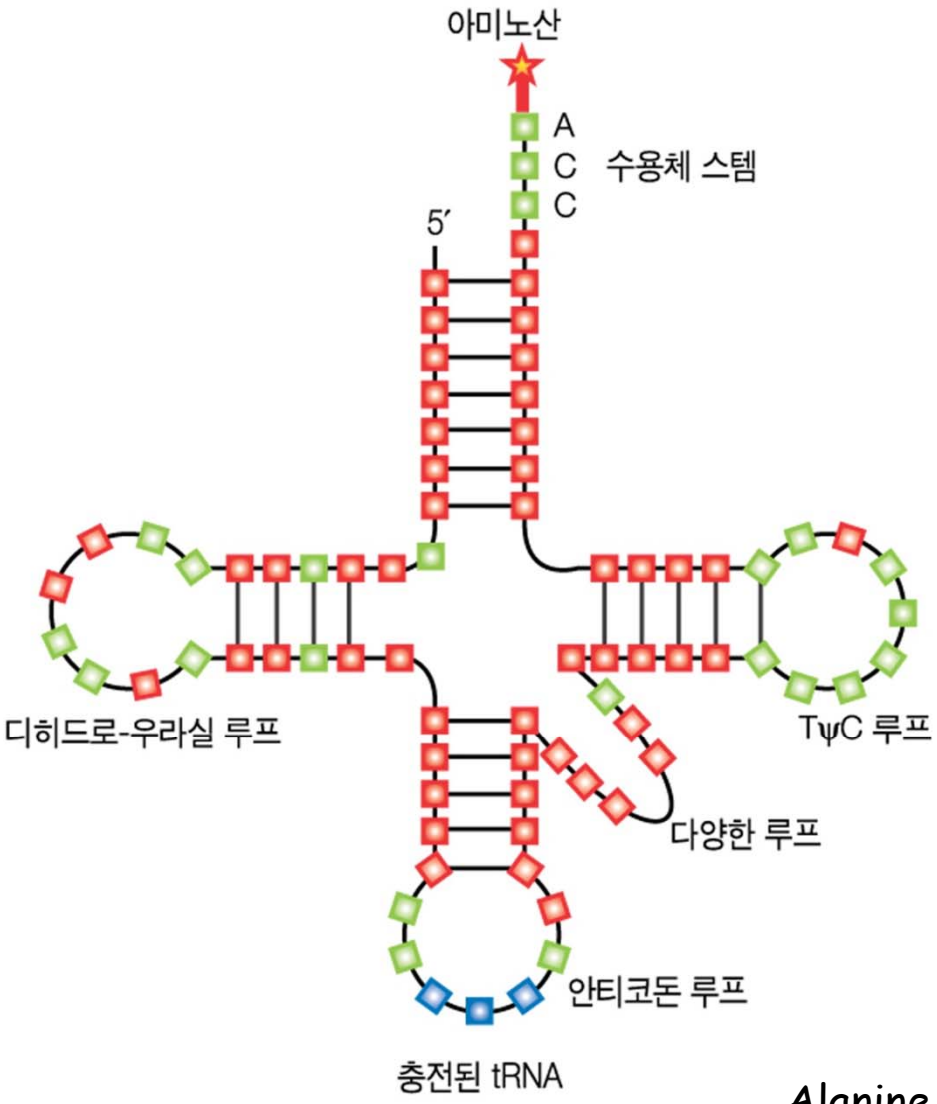


Figure 12-7 part 1 Principles of Genetics, 4/e

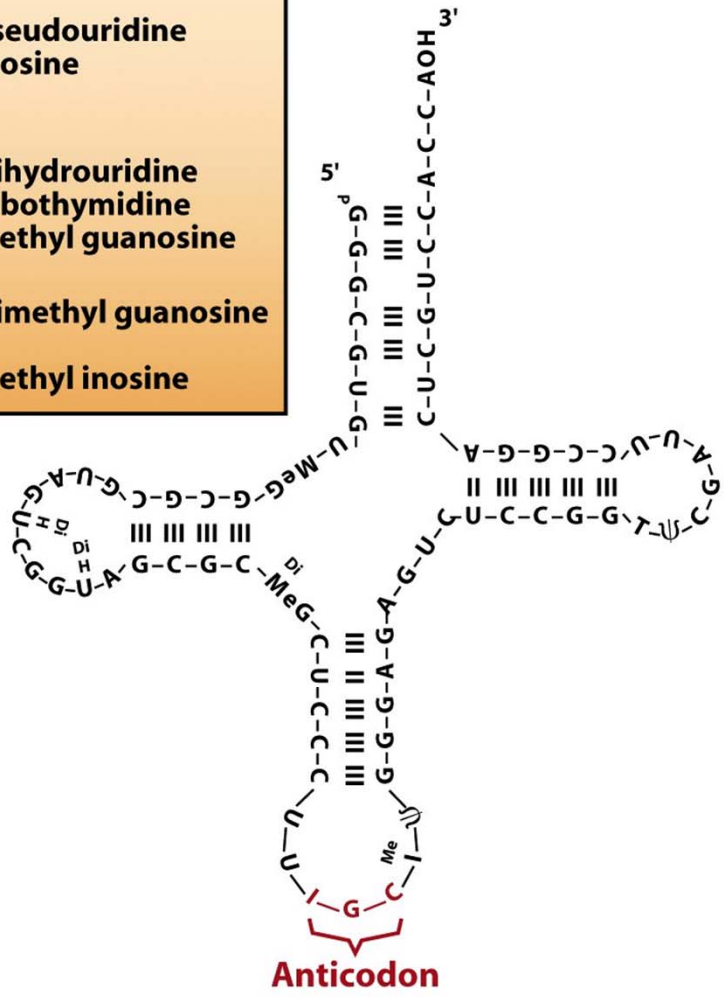
Transfer RNA

- **Transfer RNA (tRNA)** carries the AA corresponding to the codon it recognizes and move it to ribosome
- tRNA: 4S 70~95 nucleotides long
- contain a triplet nucleotide sequence, the anticodon, which is complementry to and base pairs with the codon sequence in mRNA
- There are one to four tRNAs for each of the 20 amino acids(AA)
- The amino acids are attached to the tRNAs by high-energe bonds between **carboxyl groups** of AA and **3'-hydroxyl termini** of the tRNA
- **Amino-acyl tRNA synthetases** recognizes both the tRNA and the corresponding AA and attach the AA to tRNA (charging the tRNA)

Cloverleaf Configuration of Transfer RNA(tRNA)

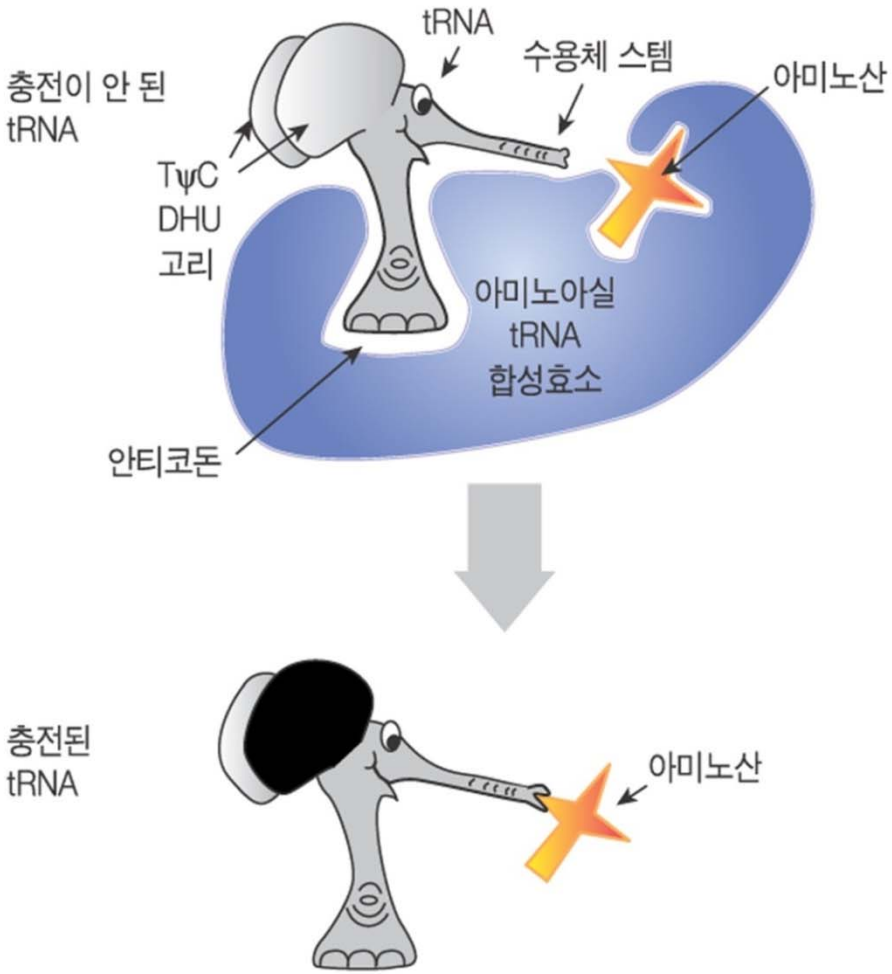


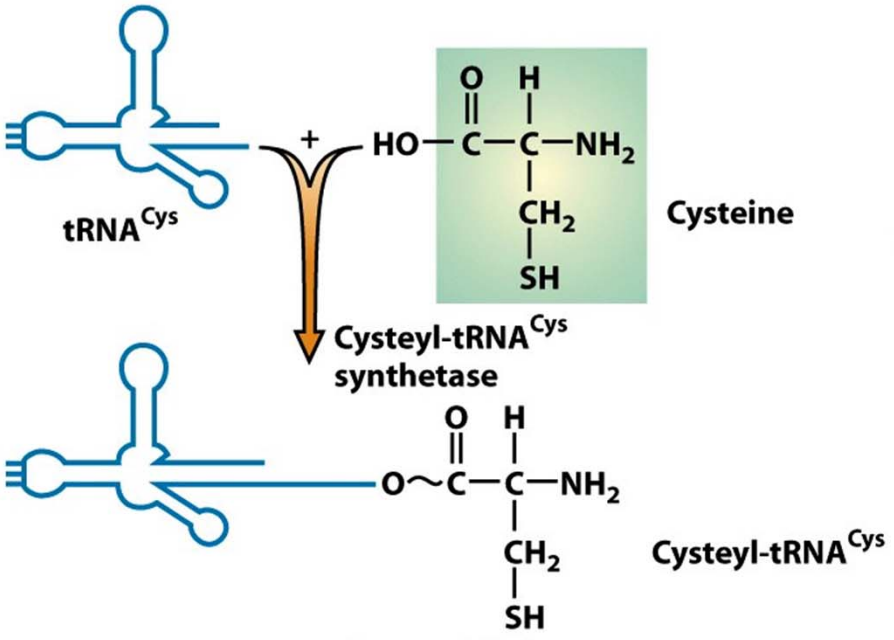
ψ	=	Pseudouridine
I	=	Inosine
DiH U	=	Dihydrouridine
T	=	Ribothymidine
MeG	=	Methyl guanosine
DiMeG	=	Dimethyl guanosine
MeI	=	Methyl inosine



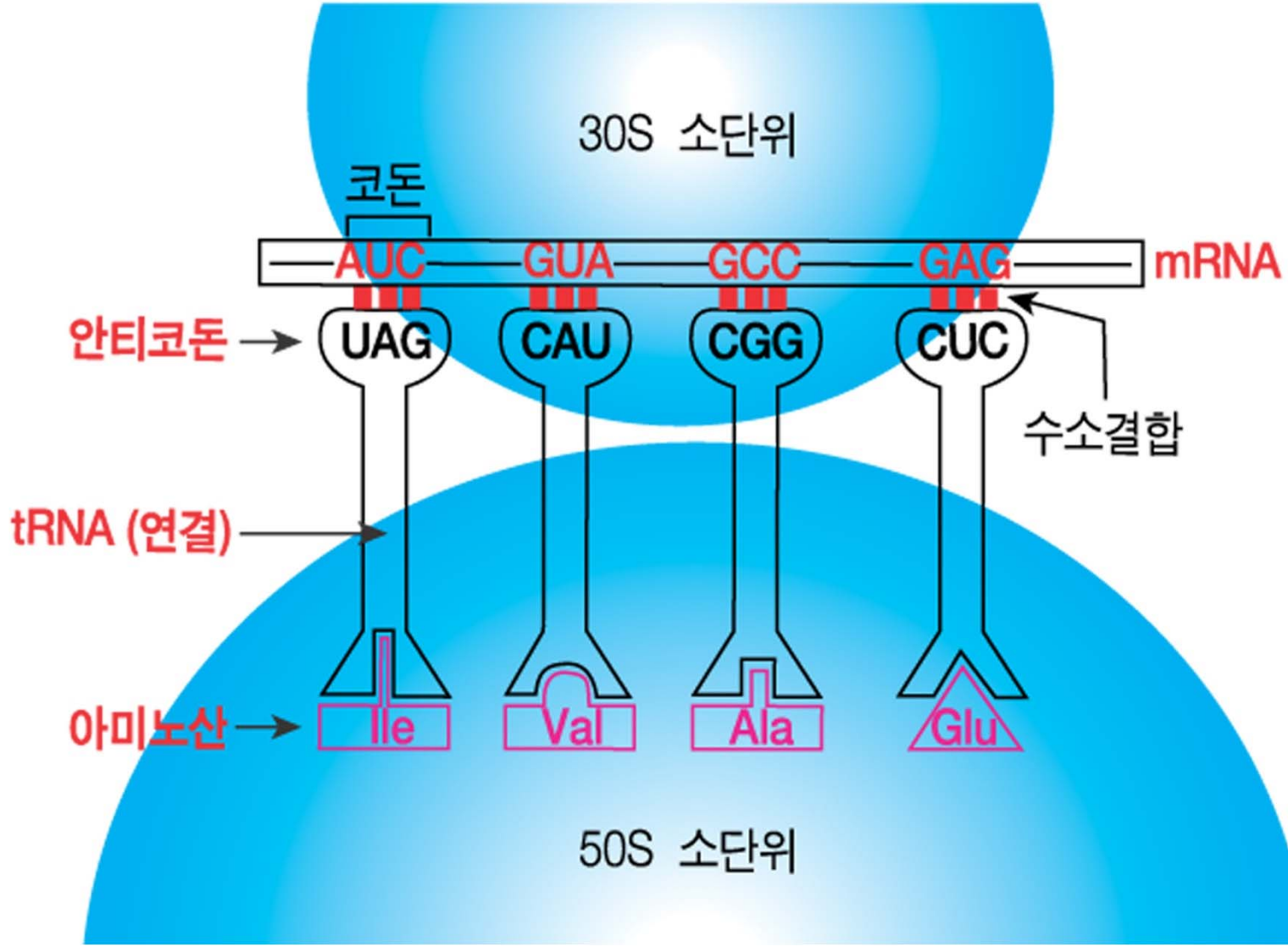
Alanine tRNA of S. cerevisiae: 1965, Robert W. Holley

Aminoacyl-tRNA formation by aminoacyl-tRNA synthetase





Codon-recognizing specificity of
an aminoacyl-tRNA complex


30S 소단위
코돈
AUC
GUA
GCC
GAG
mRNA
안티코돈
UAG
CAU
CGG
CUC
수소결합
tRNA (연결)
아미노산
Ile
Val
Ala
Glu
50S 소단위

Transfer RNA

- **Three MUSTs of tRNA**

  - must have correct anticodon sequences, so as to response to the right codons
  - must be recognized by the correct amicoacyl-tRNA synthetases, so that they are activated with the correct amino acids
  - must bind to the appropriate sites on the ribosomes to carry out their adapter function

- **Three tRNA binding sites on each ribsome**

  - ***A* or aminoacyl site:** binds the incoming aminoacyl-tRNA, the tRNA carrying the next amino acid to be added to the growing polytpeptid chain
  - **P or peptidyl site:** binds the tRNA to which the growing polypeptide is attached
  - **E or exit site:** binds the departing uncharged tRNA

70S Ribosome Diagram

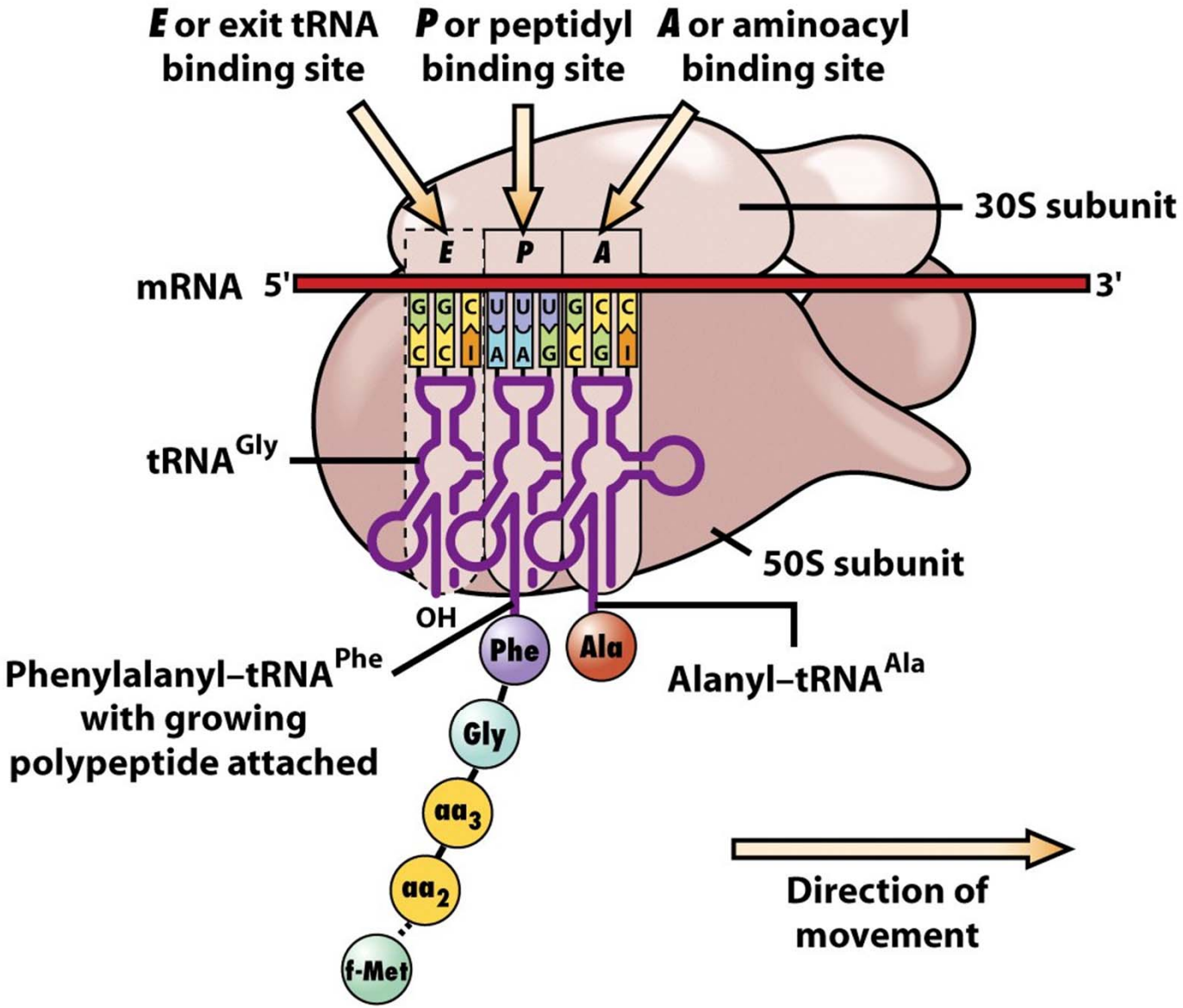


Figure 12-14a Principles of Genetics, 4/e

Process of Translation

- Ribosome binding site (**Shine-Dalgarno sequence**) near the front of mRNA
  binds to anti-S-D sequence close to 3′ end of the 16S ribosomal RNA

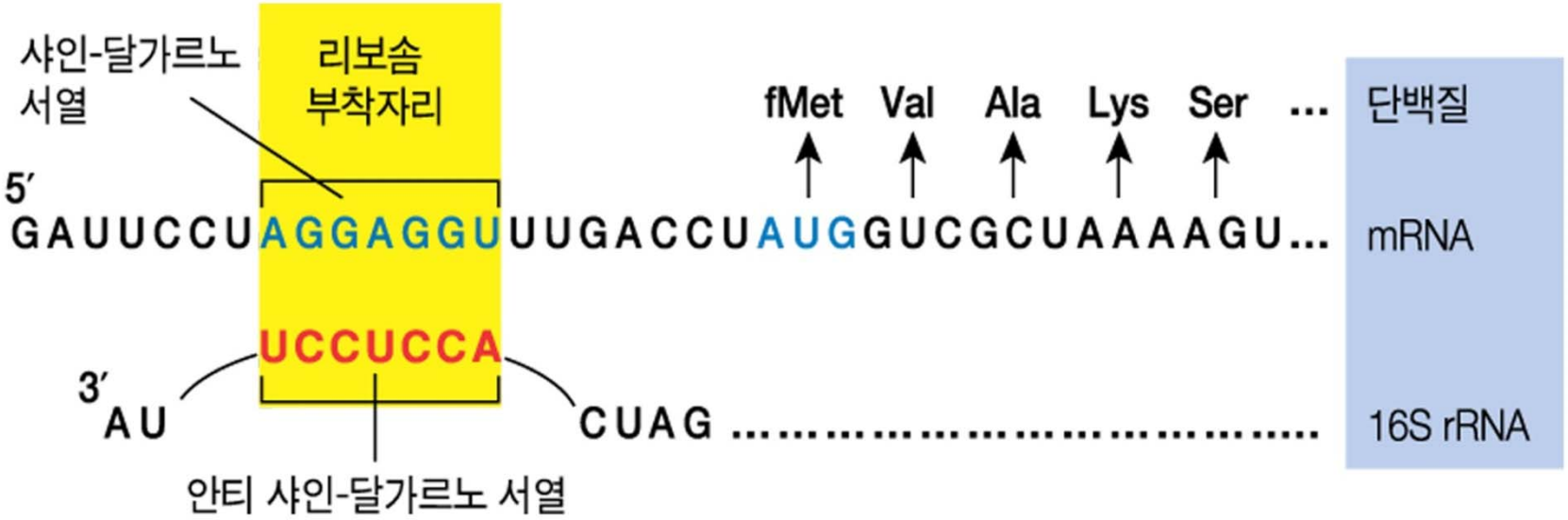
- Translation starts from **Start codon** (AUG-Met) in mRNA
  (The start codon is the next AUG codon after the ribosome binding site)

- **Open reading frame(ORF)** is a stretch of RNA, beginning with a start codon, and which can therefore be translated into protein

- Protein synthesis stops at **Stop codon**, UGA, UAG, UAA

- **Release factor** read the stop signal and chop the completed polypeptide chain off the final tRNA

Shine-Dalgarno sequence & Start codon for translation initiation



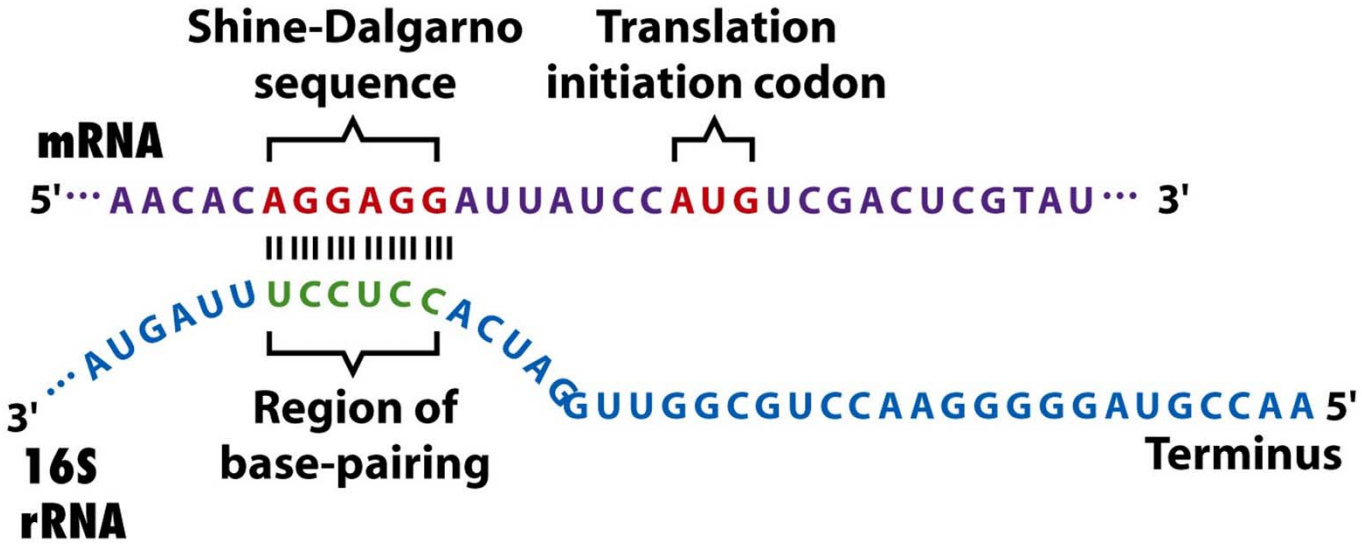
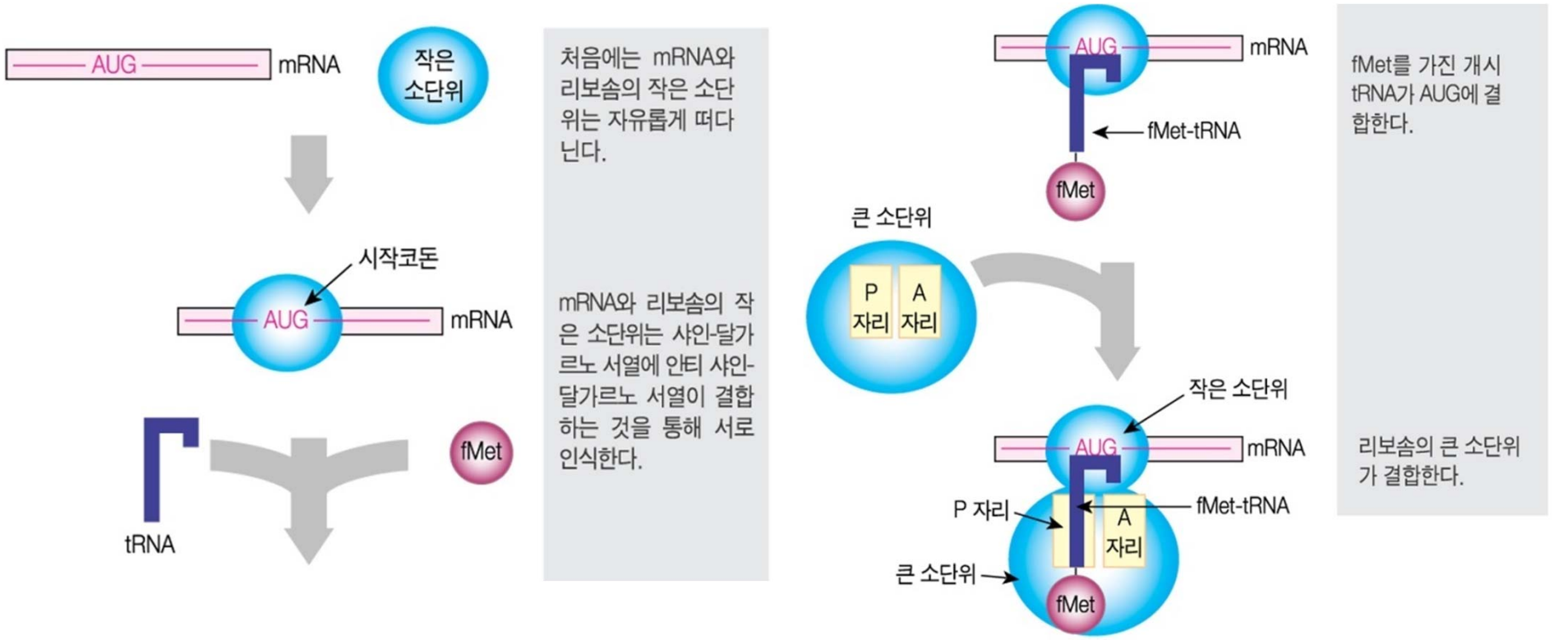
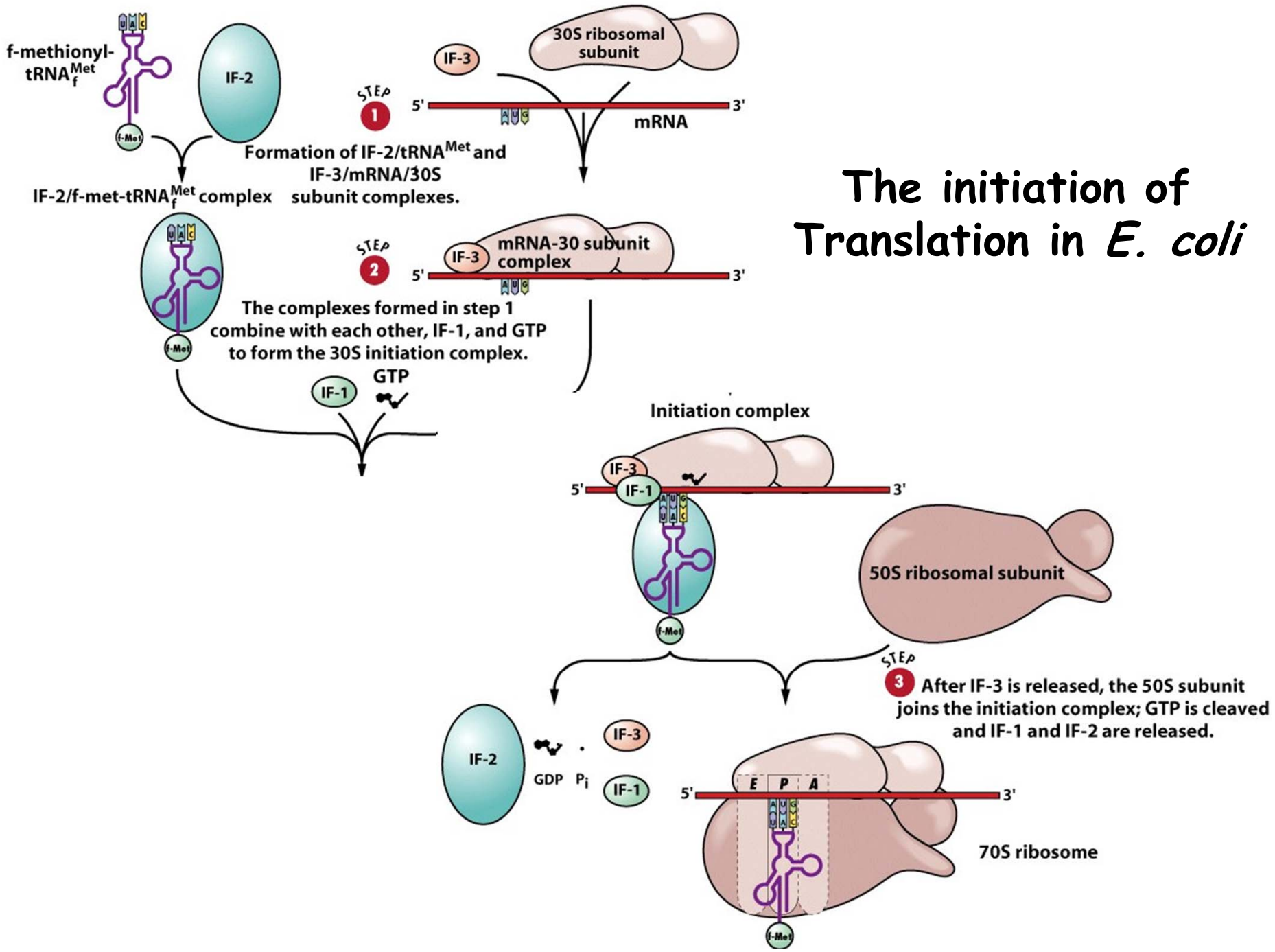


Figure 12-16 Principles of Genetics, 4/e

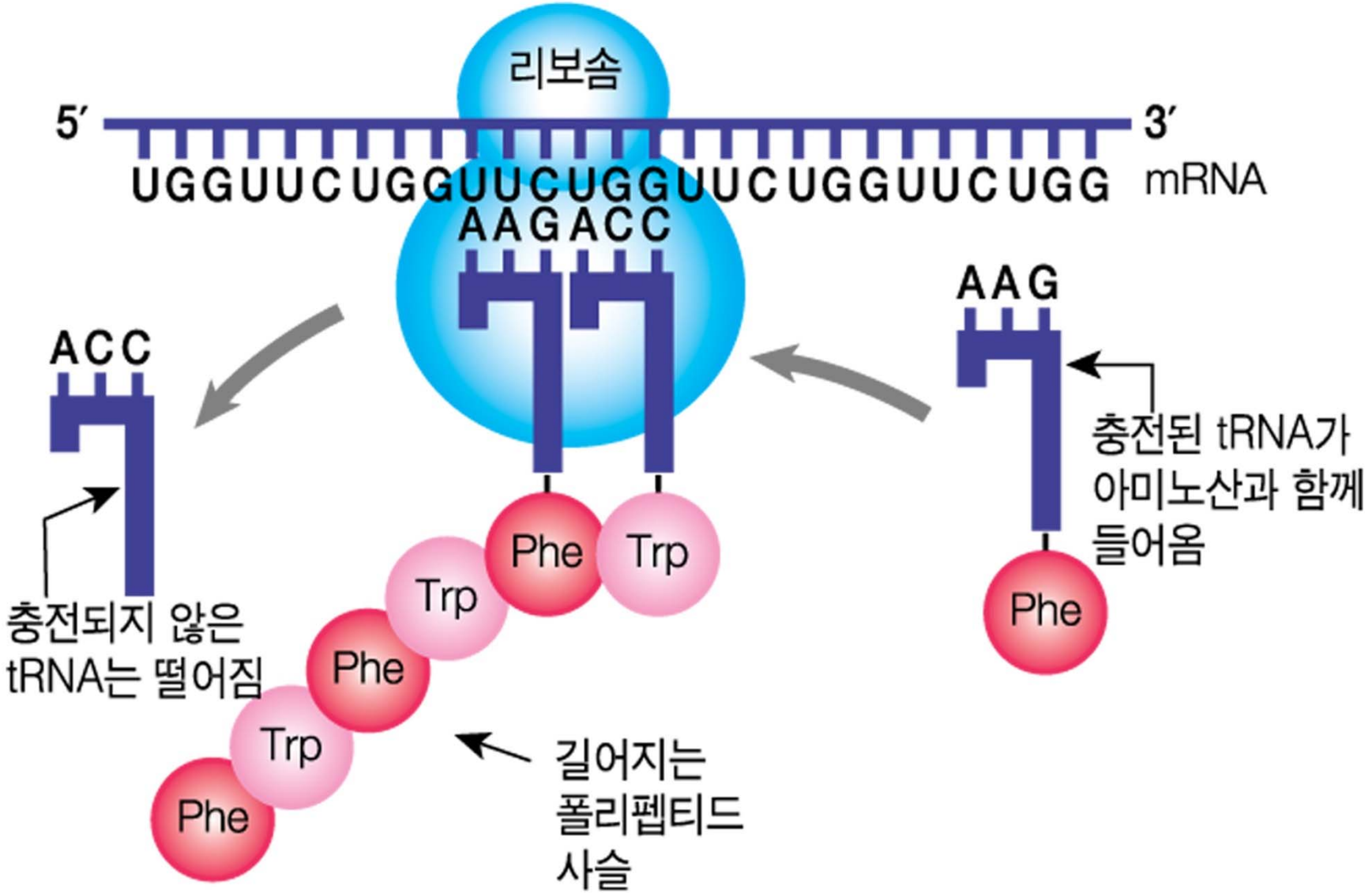
The Initiation of Translation



The initiation of Translation in *E. coli*



Elongation of polypeptide chain



Polypeptide chain elongation in *E. coli*

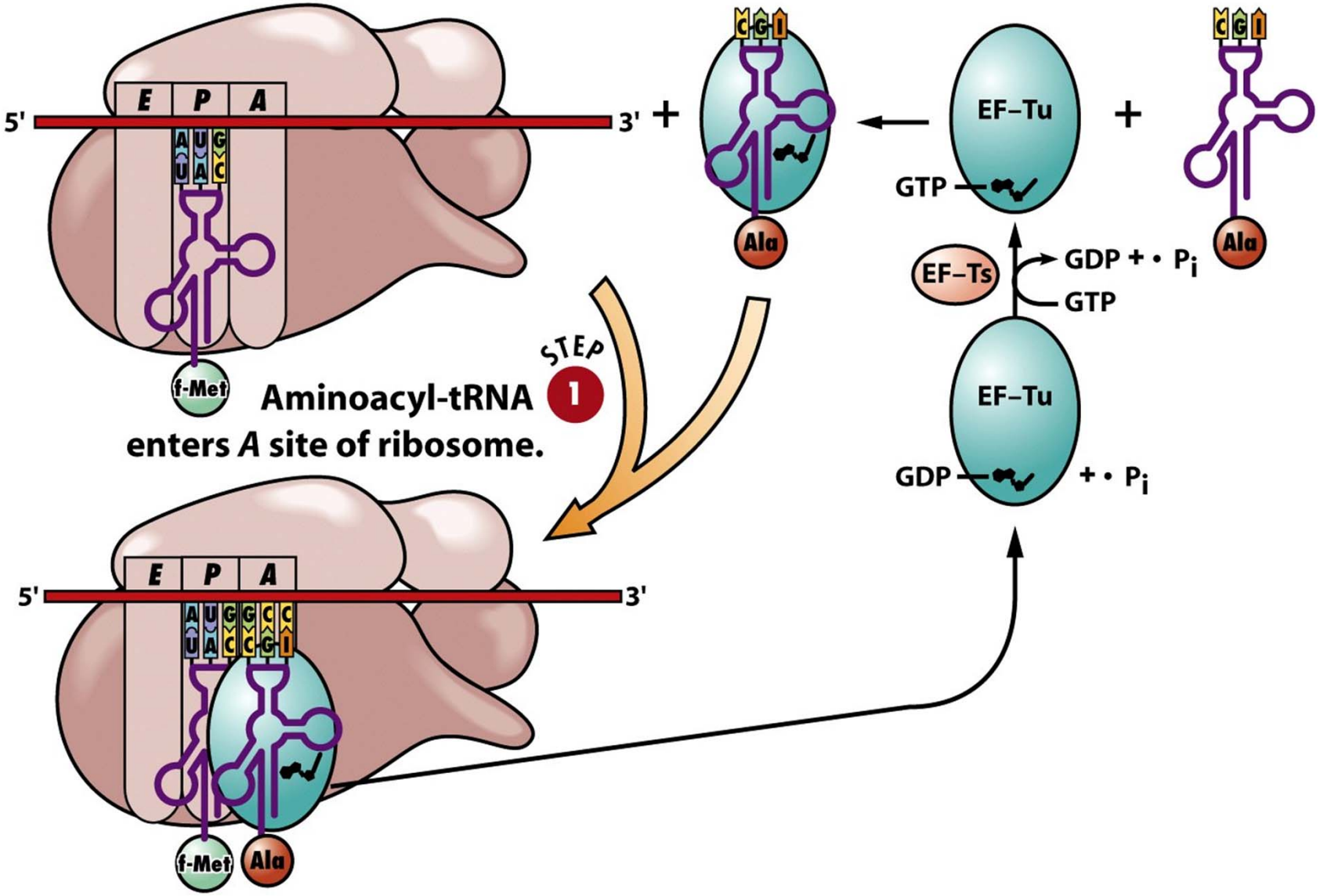


Figure 12-17 part 1 Principles of Genetics, 4/e

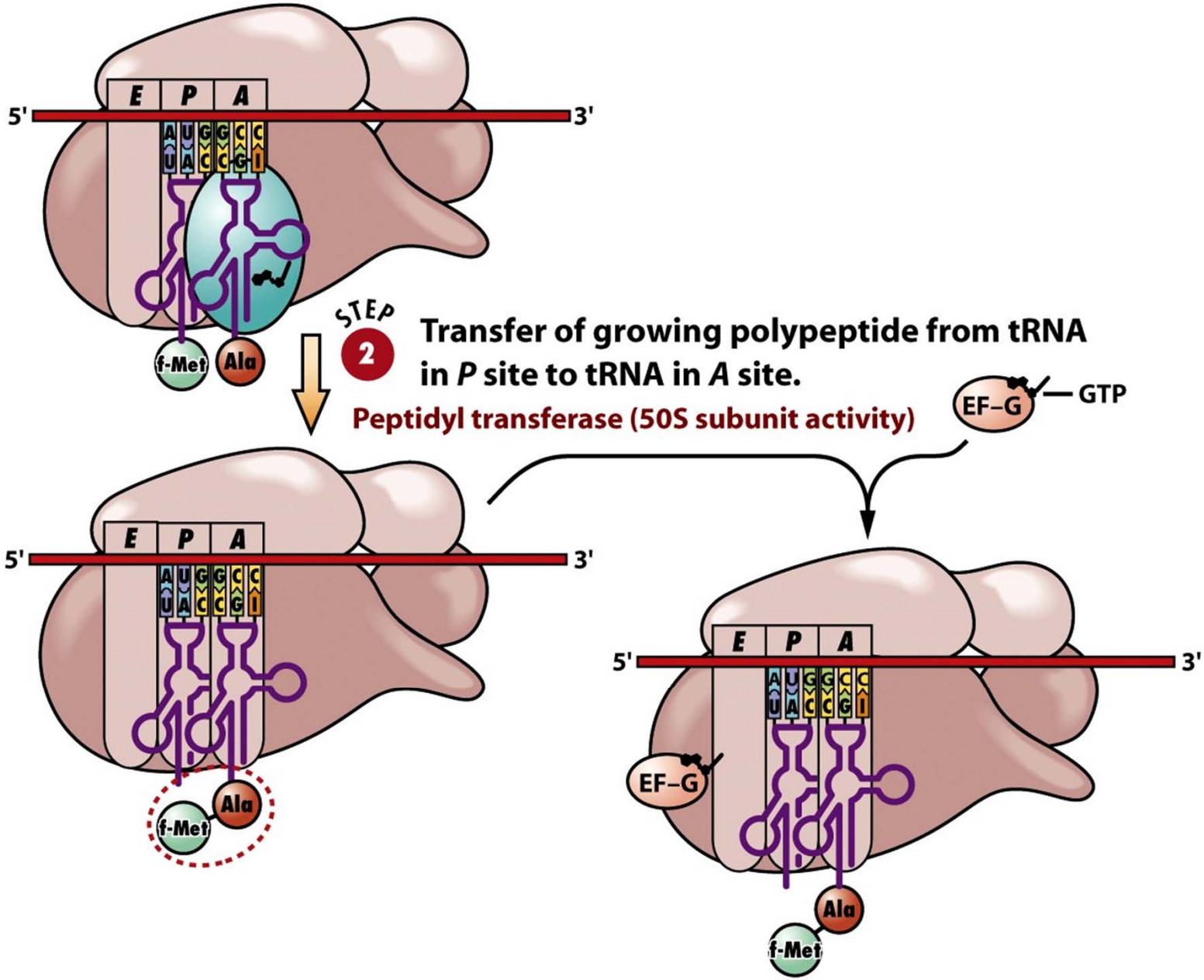


Figure 12-17 part 2 Principles of Genetics, 4/e

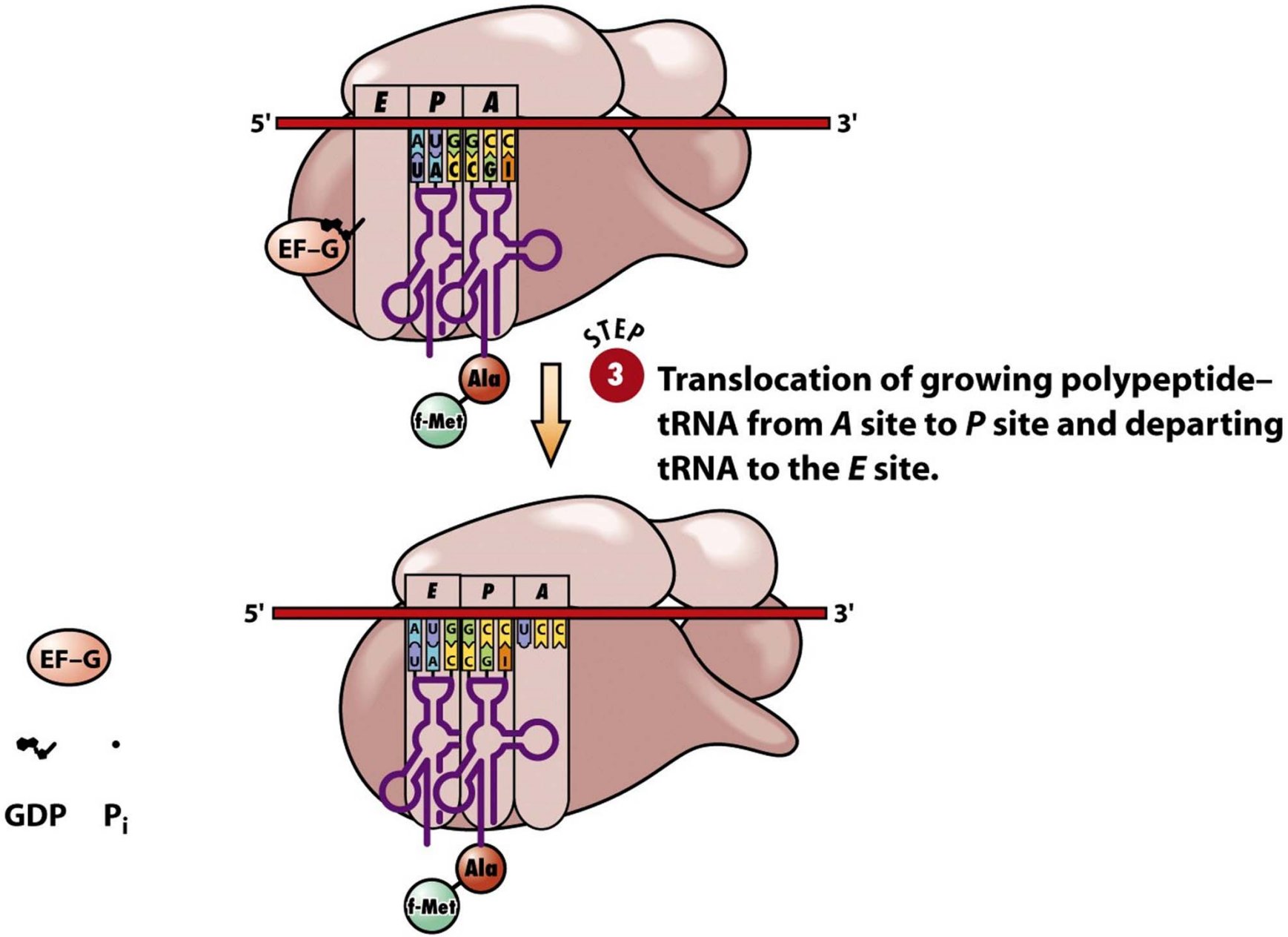


Figure 12-17 part 3 Principles of Genetics, 4/e

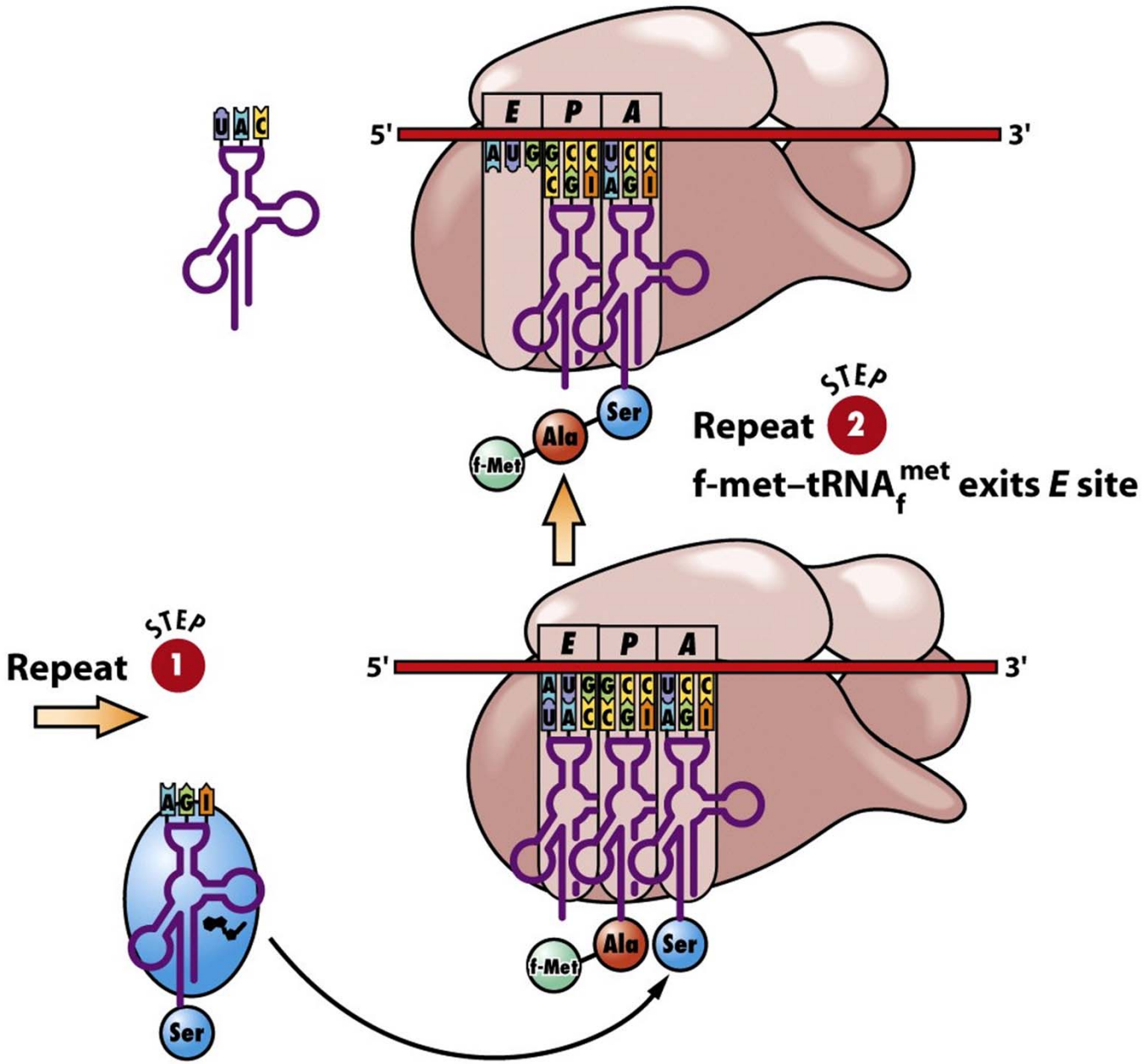


Figure 12-17 part 4 Principles of Genetics, 4/e

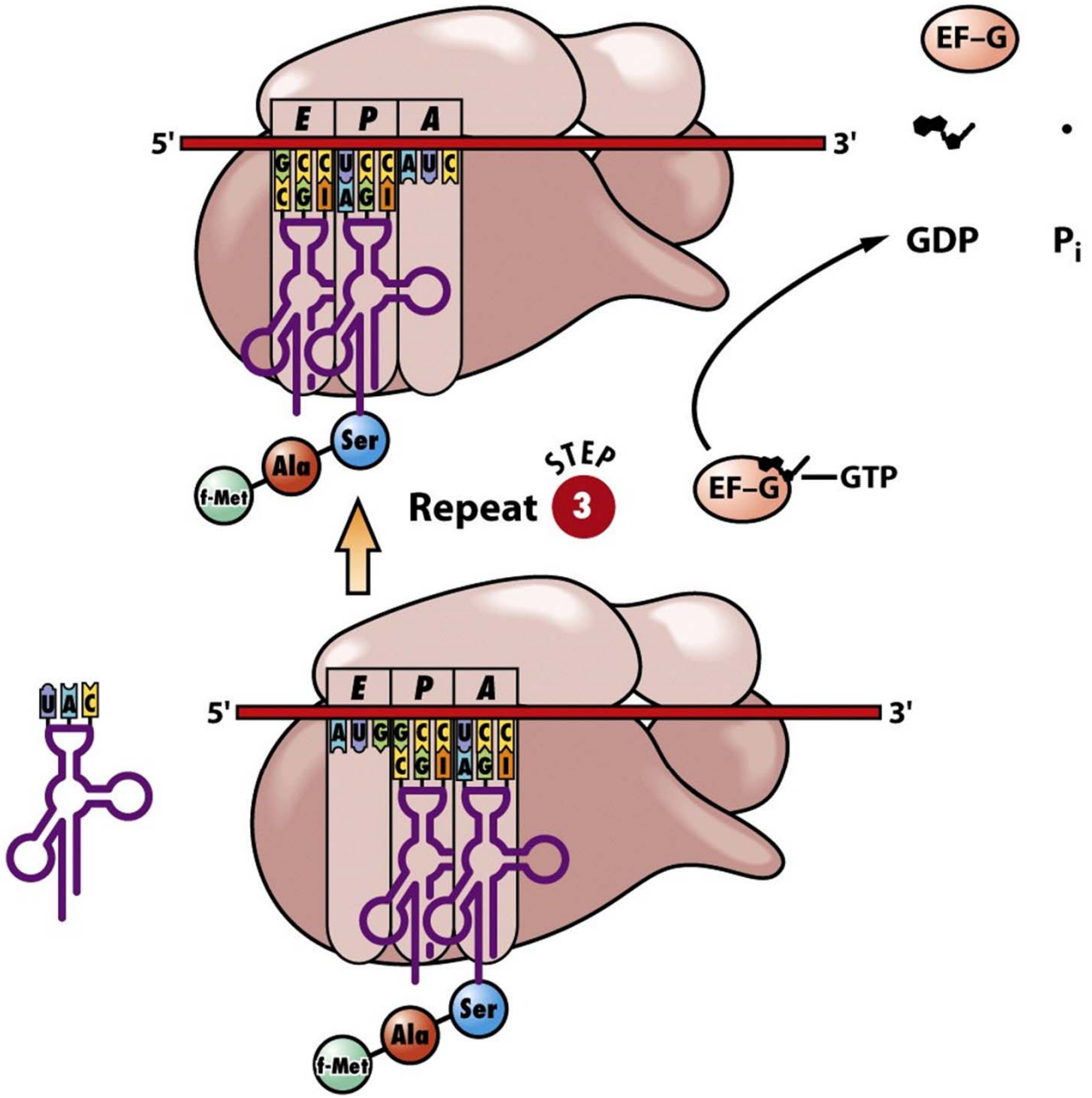


Figure 12-17 part 5 Principles of Genetics, 4/e

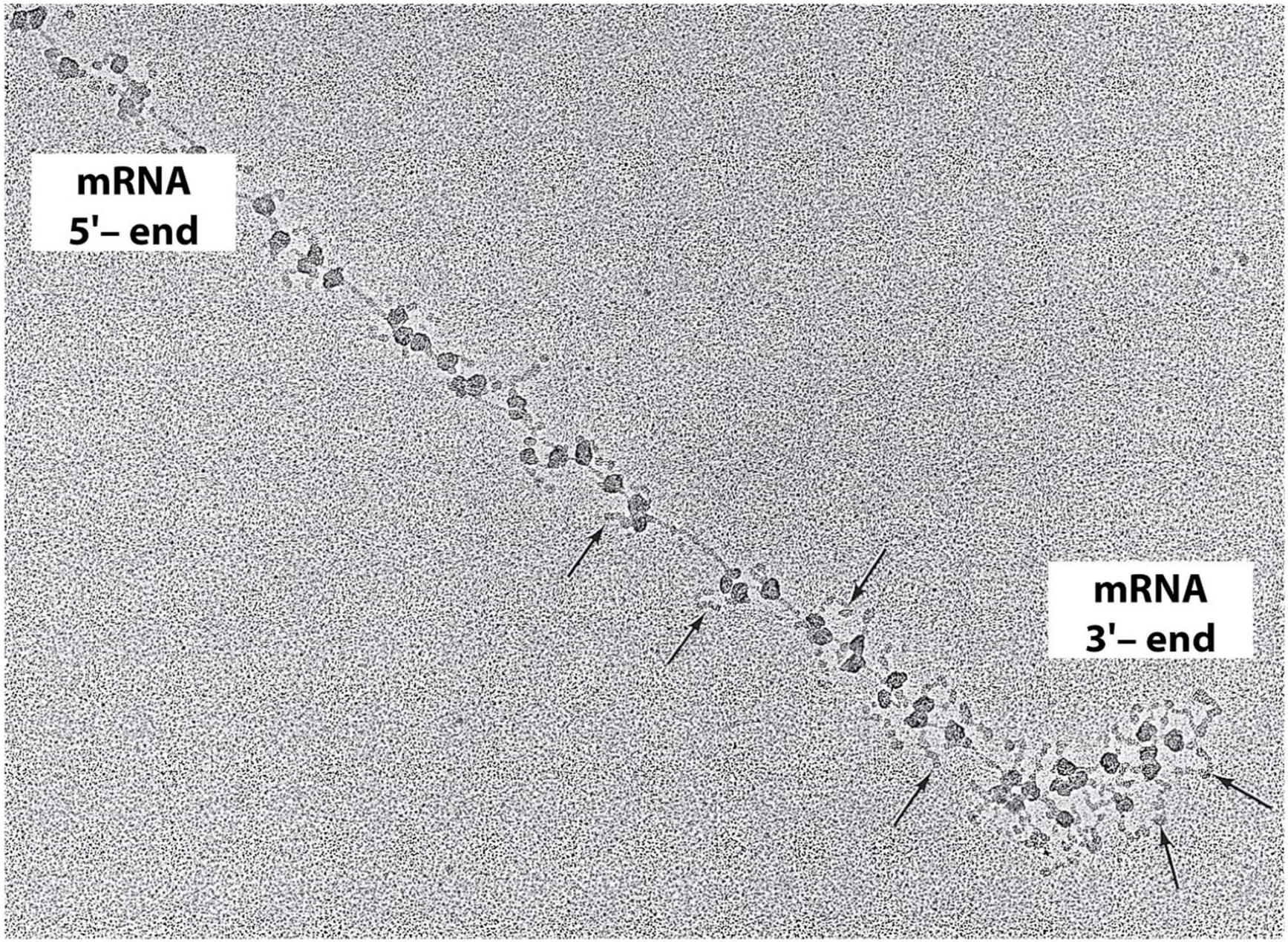
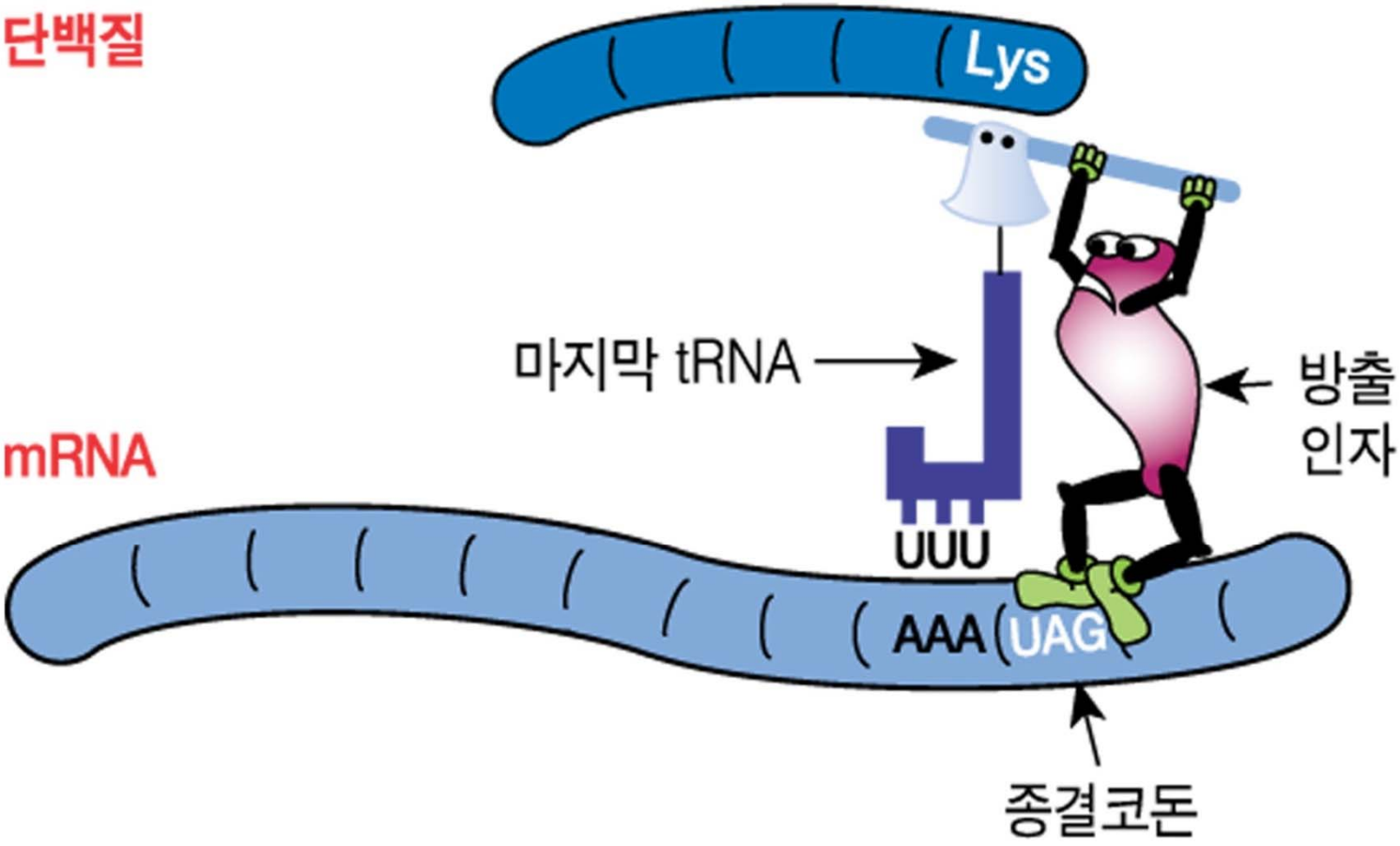


Figure 12-18 Principles of Genetics, 4/e

Termination of polypeptide chain elongation



when any of the three chain-termination codon(UAA, UAG, UGA) enters the A site on the ribosome

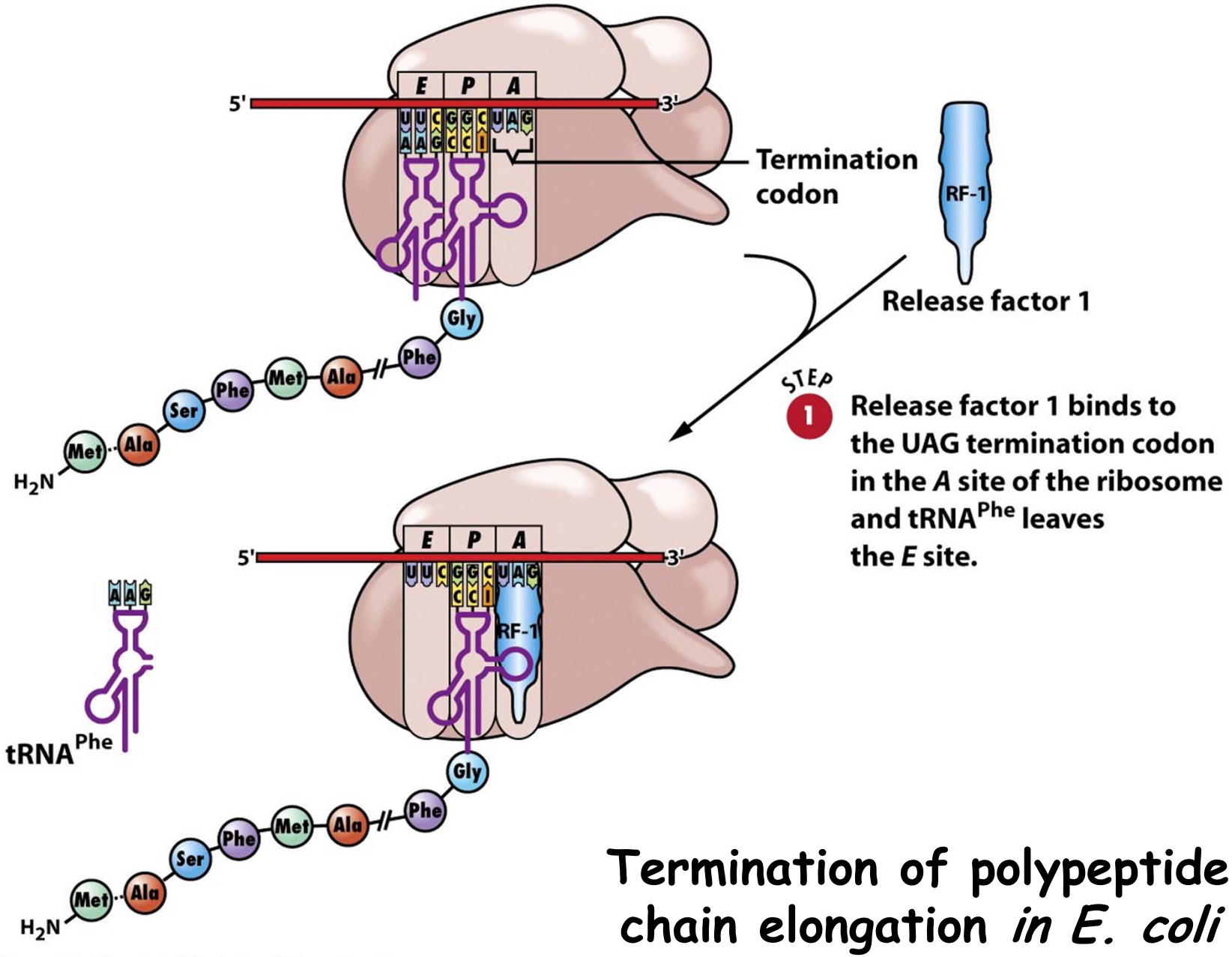


Figure 12-19 part 1 Principles of Genetics, 4/e

Termination of polypeptide chain elongation *in E. coli*

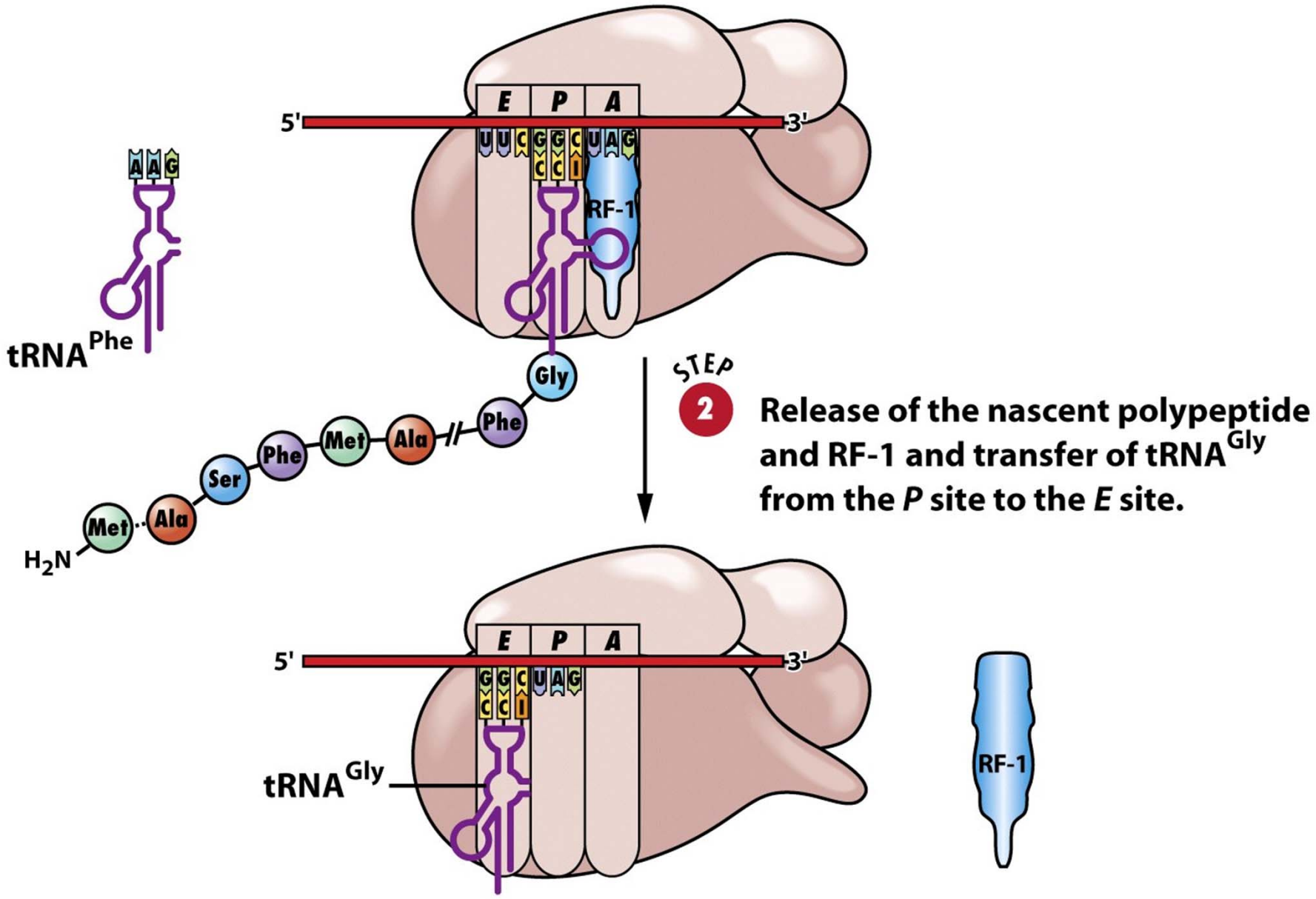


Figure 12-19 part 2 Principles of Genetics, 4/e

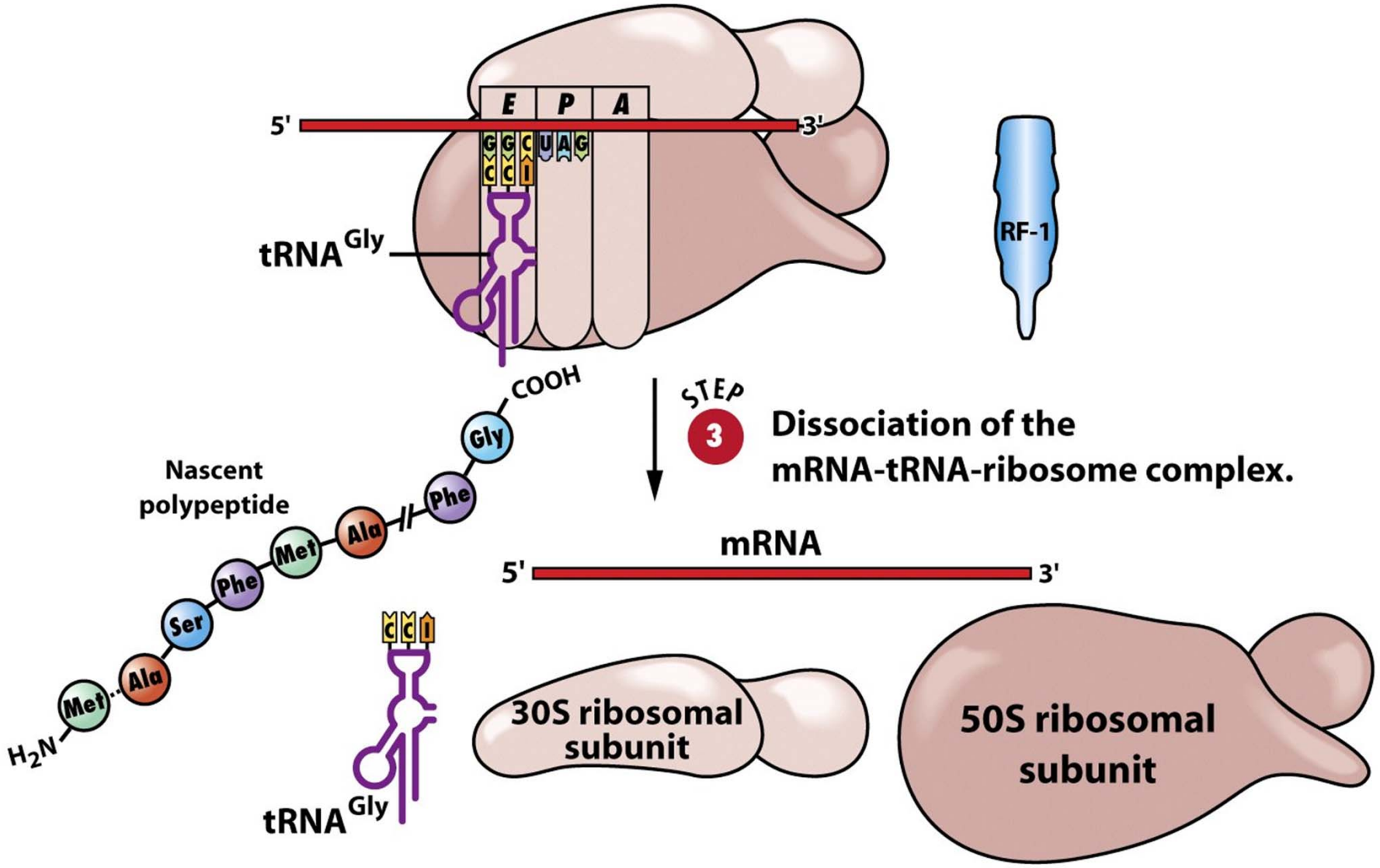


Figure 12-19 part 3 Principles of Genetics, 4/e

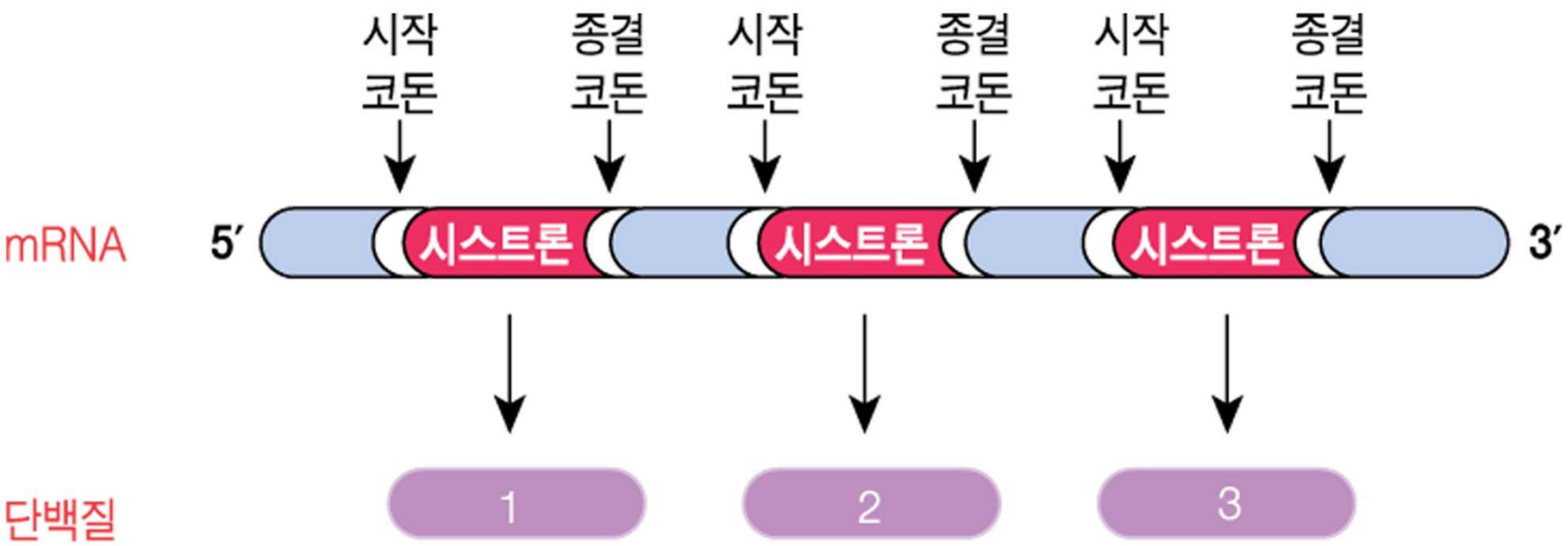
Special aspects of translation in Bacteria

- In prokaryotes, one **polycistronic mRNA** can code for several proteins

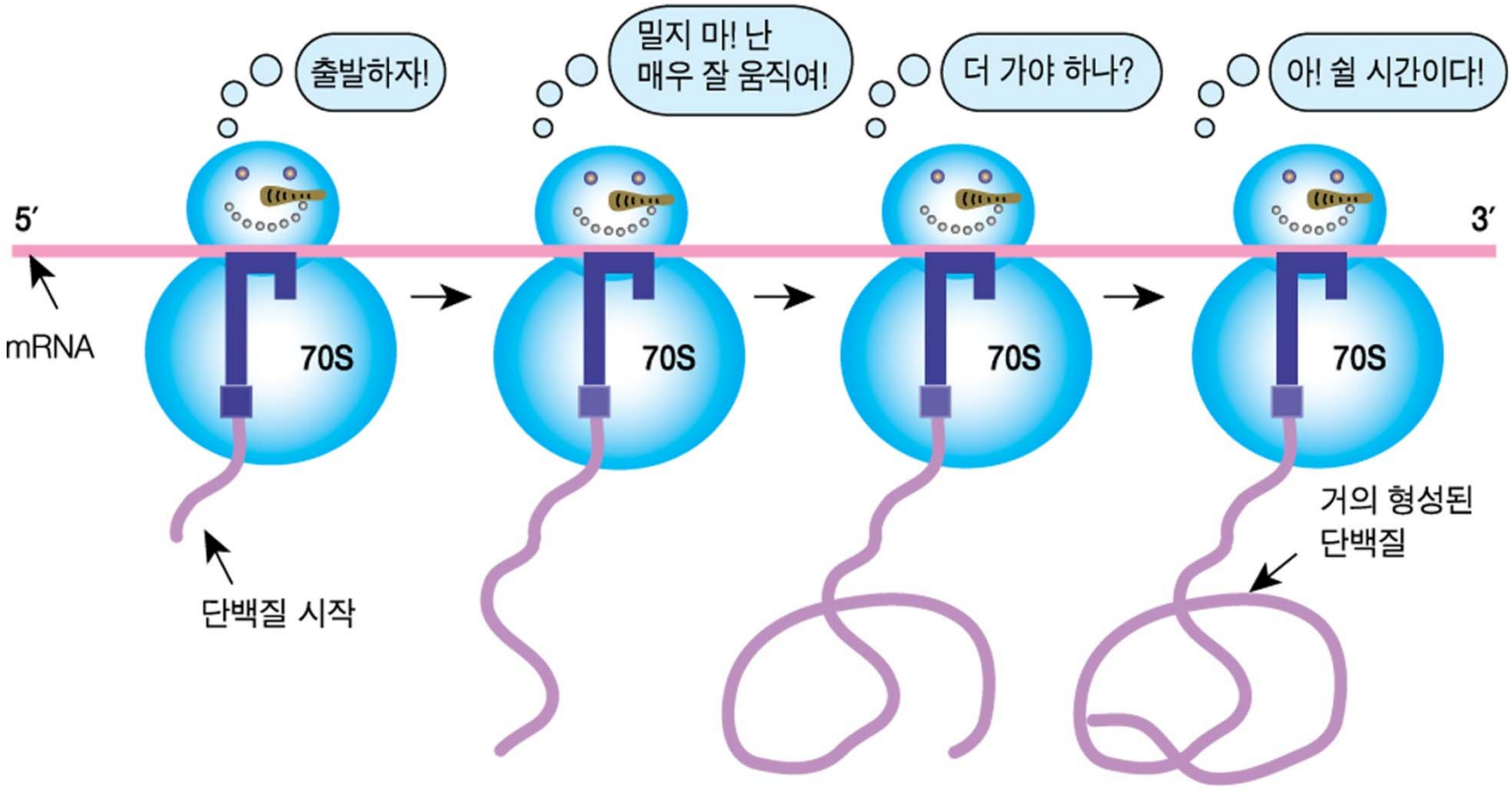
- Several ribosomes can read the same mRNA at once (**Polysome**; a group of ribosomes that bind to and translate the same mRNA)

- Translation and transcription can be coupled in Bacteria
  - **Coupled translation-transcription**; when ribosomes of bacteria start translating an mRNA molecule which is still being transcribed from the gene

Polycistronic mRNA in Bacteria



Polysome in Bacteria



Synthesize many protein molecules in short time !

Coupled translation-transcription in Bacteria