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Chapter 32: Translation

Voet & Voet:
Pages 1343-1385
(Parts of sections 1-3)

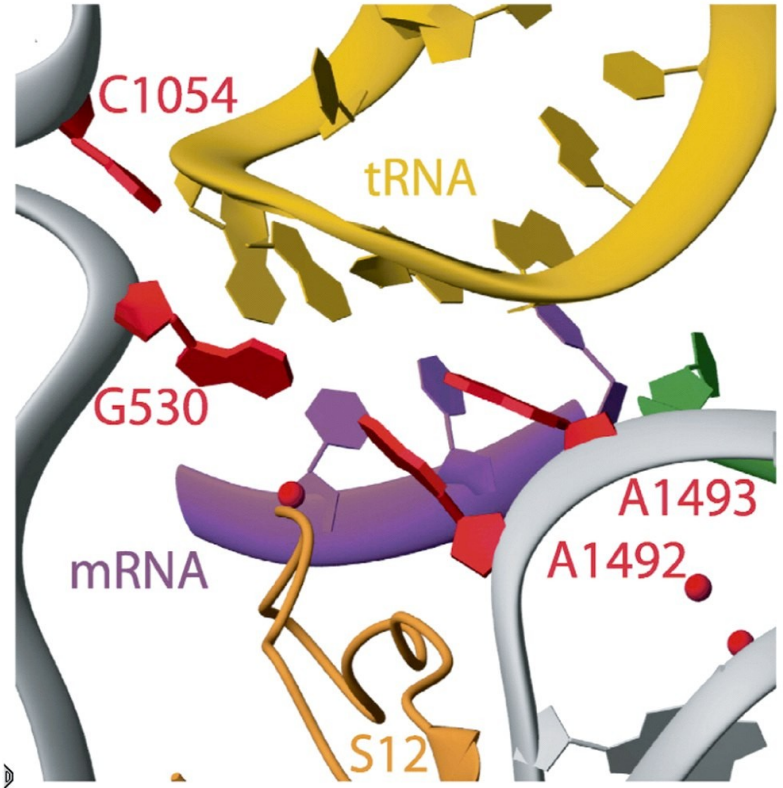

Figure 26-33b Fundamentals of Biochemistry, 2/e

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Genetic code

Translates the genetic information into functional proteins

mRNA is read in 5‘ to 3‘ direction

Codons are base triplets representing one amino acid

e.g. GCA = Ala

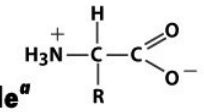

Table 26-1. Key to Function. The "Standard" Genetic Code[a]

First position (5′ end)	Second position U	Second position C	Second position A	Second position G	Third position (3′ end)
U	UUU Phe	UCU	UAU Tyr	UGU Cys	U
	UUC Phe	UCC	UAC Tyr	UGC Cys	C
	UUA Leu	UCA Ser	UAA STOP	UGA STOP	A
	UUG Leu	UCG Ser	UAG STOP	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 Ile	ACU Thr	AAU Asn	AGU Ser	U
	AUC Ile	ACC Thr	AAC Asn	AGC Ser	C
	AUA Ile	ACA Thr	AAA Lys	AGA Arg	A
	AUG Met[b]	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

[a] Nonpolar amino acid residues are tan, basic residues are blue, acidic residues are red, and polar uncharged residues are purple

[b] AUG forms part of the initiation signal as well as coding for internal Met residues.

Table 26-1 Fundamentals of Biochemistry, 2/e

Four features of the genetic code

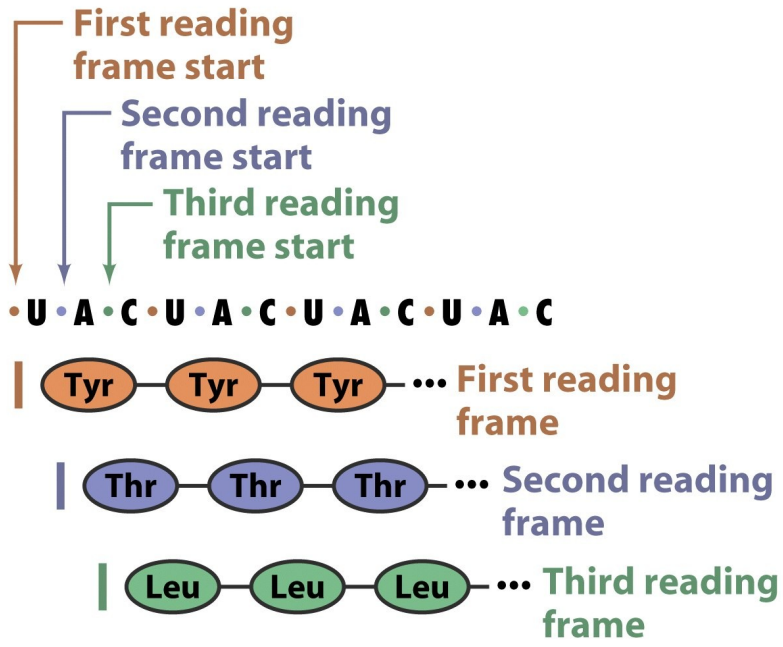

Figure 26-2 Fundamentals of Biochemistry, 2/e

TABLE 27–3 Degeneracy of the Genetic Code

Amino acid	*Number of codons*	*Amino acid*	*Number of codons*
Met	1	Tyr	2
Trp	1	Ile	3
Asn	2	Ala	4
Asp	2	Gly	4
Cys	2	Pro	4
Gln	2	Thr	4
Glu	2	Val	4
His	2	Arg	6
Lys	2	Leu	6
Phe	2	Ser	6

1. Read in a particular **frame**,
⇒ Starting point determines amino acid sequence!

2. **Degenerate**: 1 amino acid can be encoded by up to 6 codons

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Genetic code - Features 2

3. Non random:

similar amino acids are encoded by similar codons

⇒Change of 1 base has only small effect on protein

4. Widespread, almost universal:

same in most organisms from bacteria to human with very few exceptions

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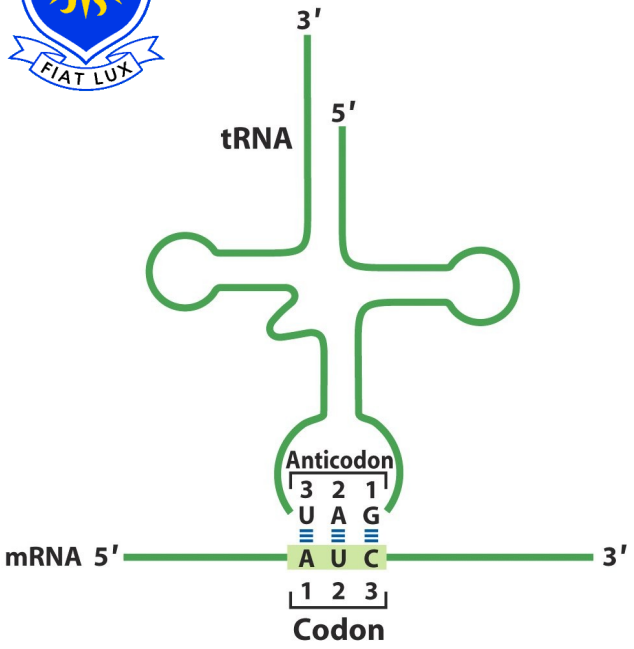
	UUG Leu	UCG	UAG STOP	UGG Trp	G
C	CUU	CCU	CAU His	CGU	U
	CUC	CCC	CAC His	CGC	C
	CUA Leu	CCA Pro	CAA Gln	CGA Arg	A
	CUG	CCG	CAG Gln	CGG Arg	G
A	AUU	ACU	AAU Asn	AGU Ser	U
	AUC Ile	ACC	AAC Asn	AGC Ser	C
	AUA Ile	ACA Thr	AAA Lys	AGA	A
	AUG Met[b]	ACG Thr	AAG Lys	AGG Arg	G
G	GUU	GCU	GAU Asp	GGU	U
	GUC	GCC	GAC Asp	GGC	C
	GUA Val	GCA Ala	GAA Glu	GGA Gly	A
	GUG Val	GCG Ala	GAG Glu	GGG Gly	G

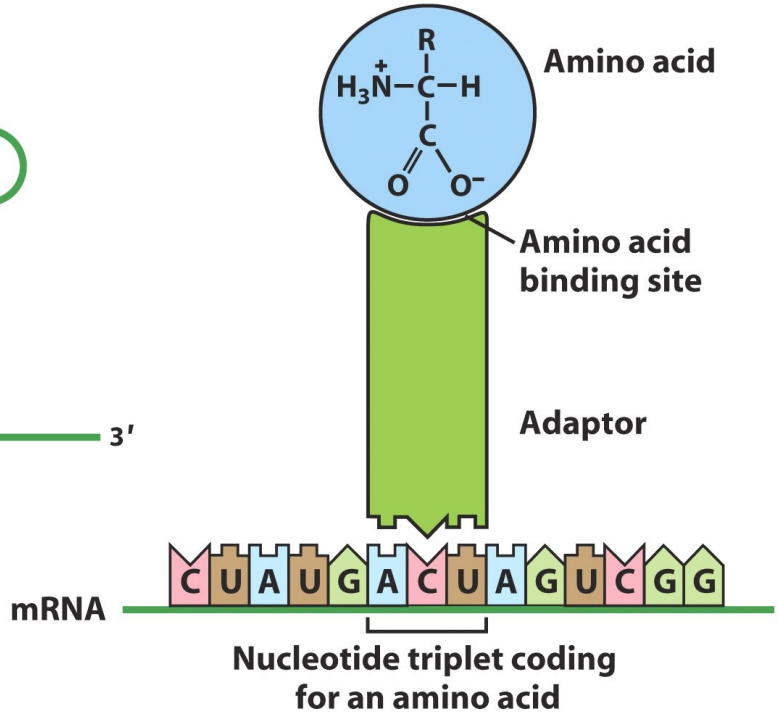
[a] Nonpolar amino acid residues are tan, basic residues are blue, acidic residues are red, and polar uncharged residues are purple

[b] AUG forms part of the initiation signal as well as coding for internal Met residues.

Table 26-1 Fundamentals of Biochemistry, 2/e

tRNAs as adaptors





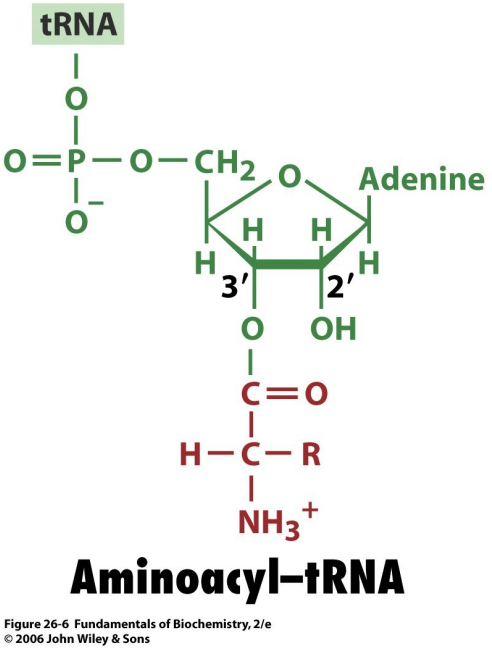


Figure 26-6 Fundamentals of Biochemistry, 2/e
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tRNA anticodon can form base pairs with mRNA codon

At least 1 tRNA for 1 amino acid

3' end of tRNA is covalently linked to amino acid

⇒Aminoacyl-tRNA

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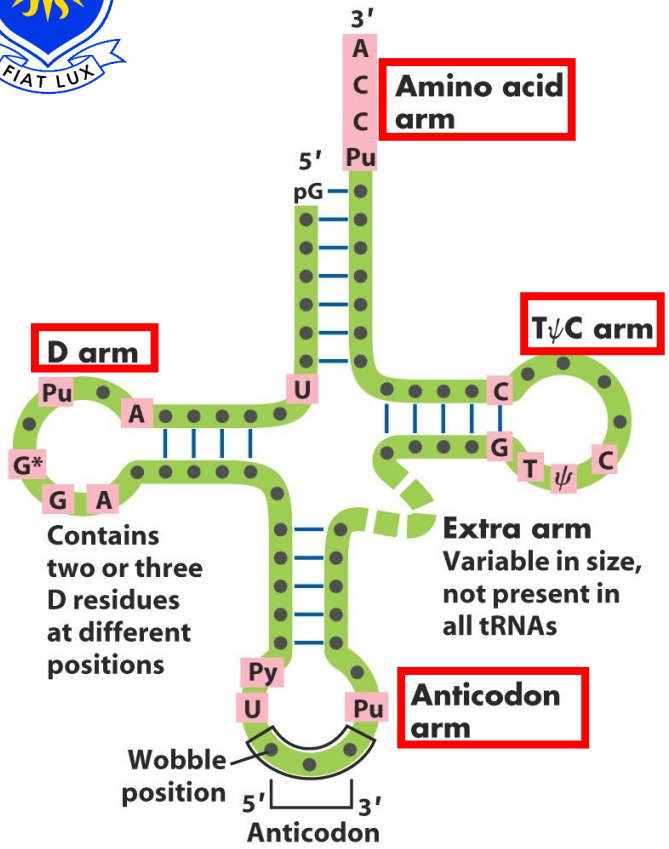
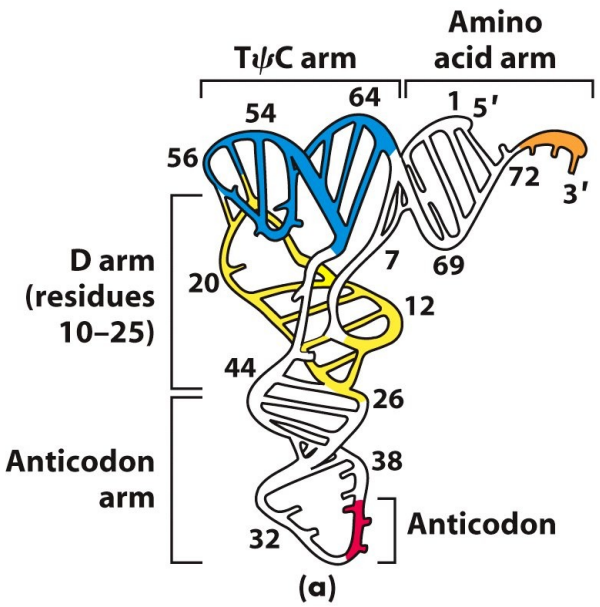
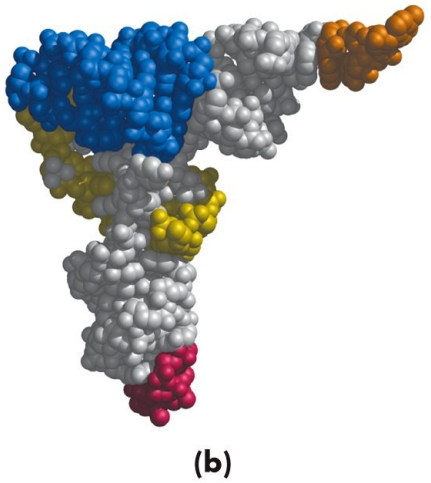
tRNA structure







L-shaped !!!

Secondary structure (base pairs):

Cloverleaf structure

Aminoacyl-end and anticodon are on opposite sites of the tRNA.

Aminoacyl-tRNA Synthases

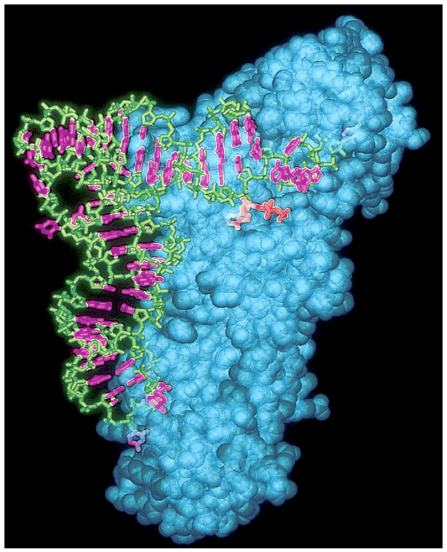
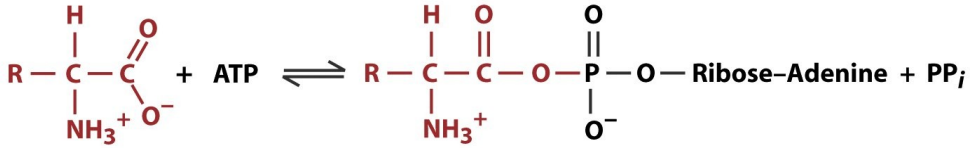


Figure 26-9 Fundamentals of Biochemistry, 2/e

One or more enzymes for each amino acid -
Specifically recognize tRNA & attach amino acid to its 3‘end:

1. Activation of amino acid by forming aminoacyl-AMP:



Amino acid **Aminoacyl–adenylate (aminoacyl–AMP)**

Unnumbered figure pg 969 Fundamentals of Biochemistry, 2/e
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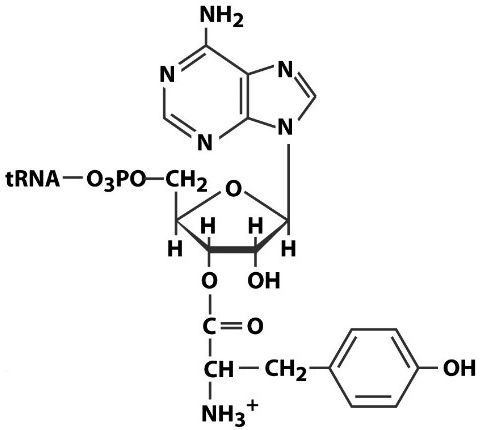
2. Transfer of amino acid to tRNA‘s 2‘ or 3‘ OH (ribose) by forming an ester bond with carboxyl group of amino acid



Tyrosyl–tRNA

3. Proofreading: bonds to incorrect amino acids are hydrolyzed

Wobble Hypothesis

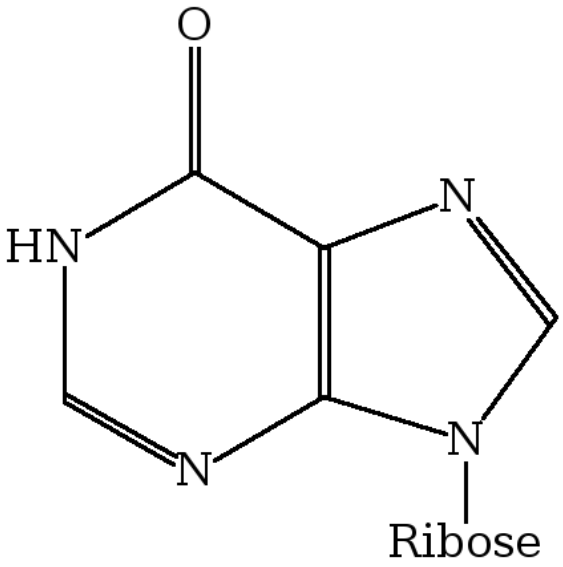
Problem: 61 sense codons versus ~ 40 different tRNAs

Solution: 3rd codon position allows non Watson-Crick base pairs

- **wobble base pairs**

Allowed wobble pairings:

5‘ anticodon	3‘ codon
C	G
A	U
U	A or G
G	U or C
I	U, C, or A



Inosine

⇒1 tRNA can recognize 1-3 codons

Procaryotic Ribosome

mRNA

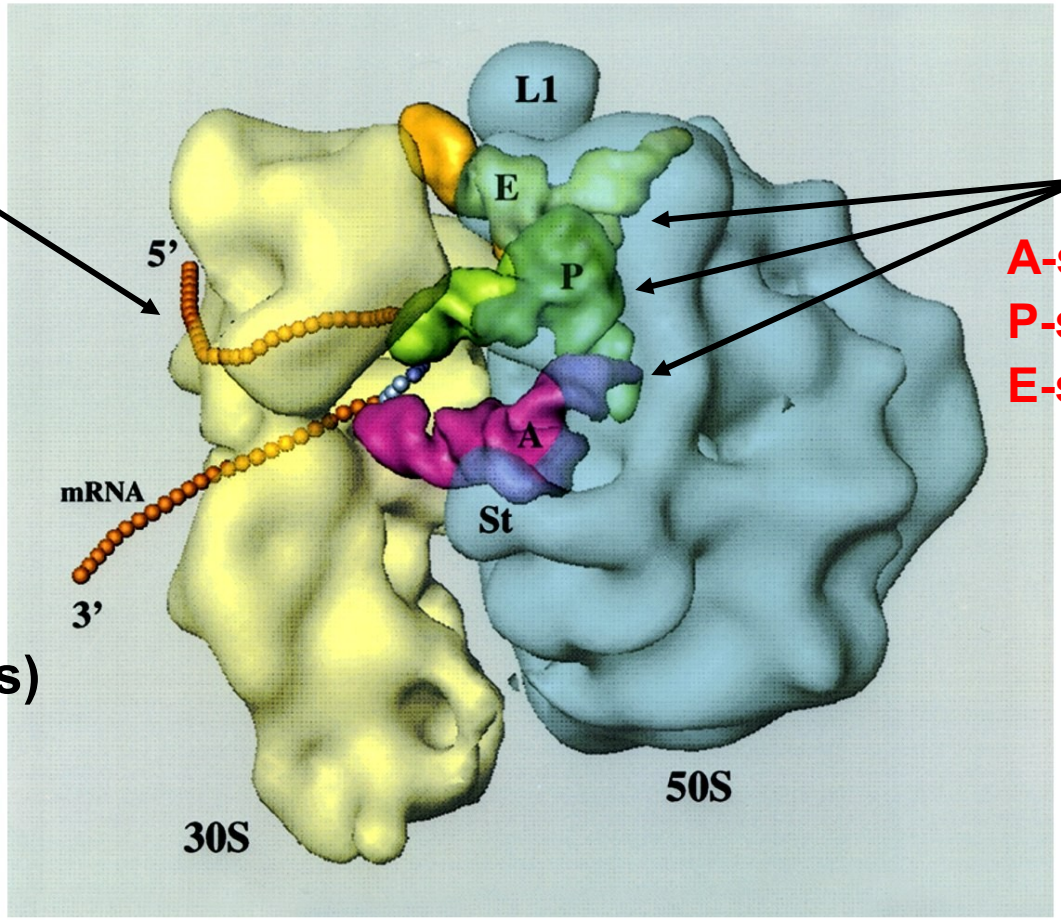


Figure 26-15b Fundamentals of Biochemistry, 2/e

tRNAs

A-site: aminoacyl tRNA

P-site: peptidyl tRNA

E-site: exit deacylated tRNA

Small Subunit (30S in prokaryotes)

Large Subunit (50S in prokaryotes)

Low-resolution structure from cryo-electron microscopy

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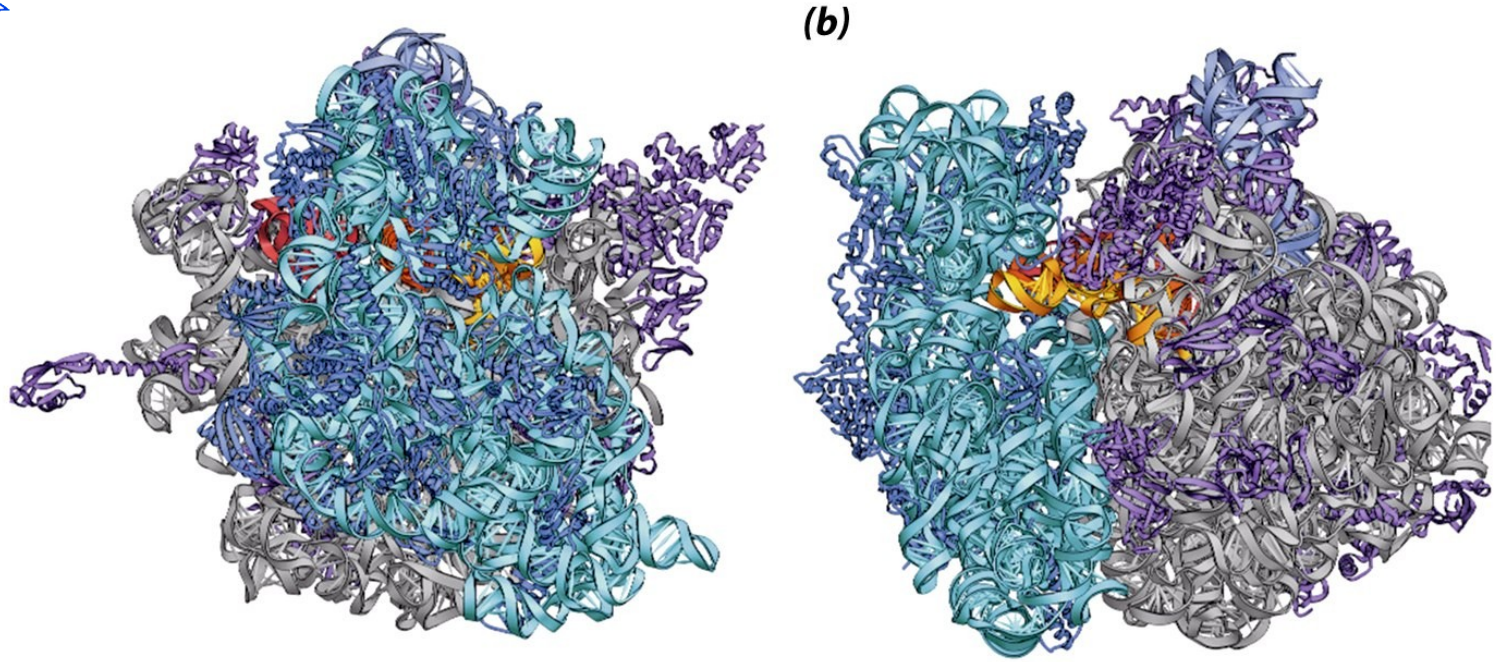
Ribosome structure



High-resolution crystal structures available since 2000

Inner core of the ribosome is RNA
Proteins 'decorate' the outer surface
=> The Ribosome is a Ribozyme!

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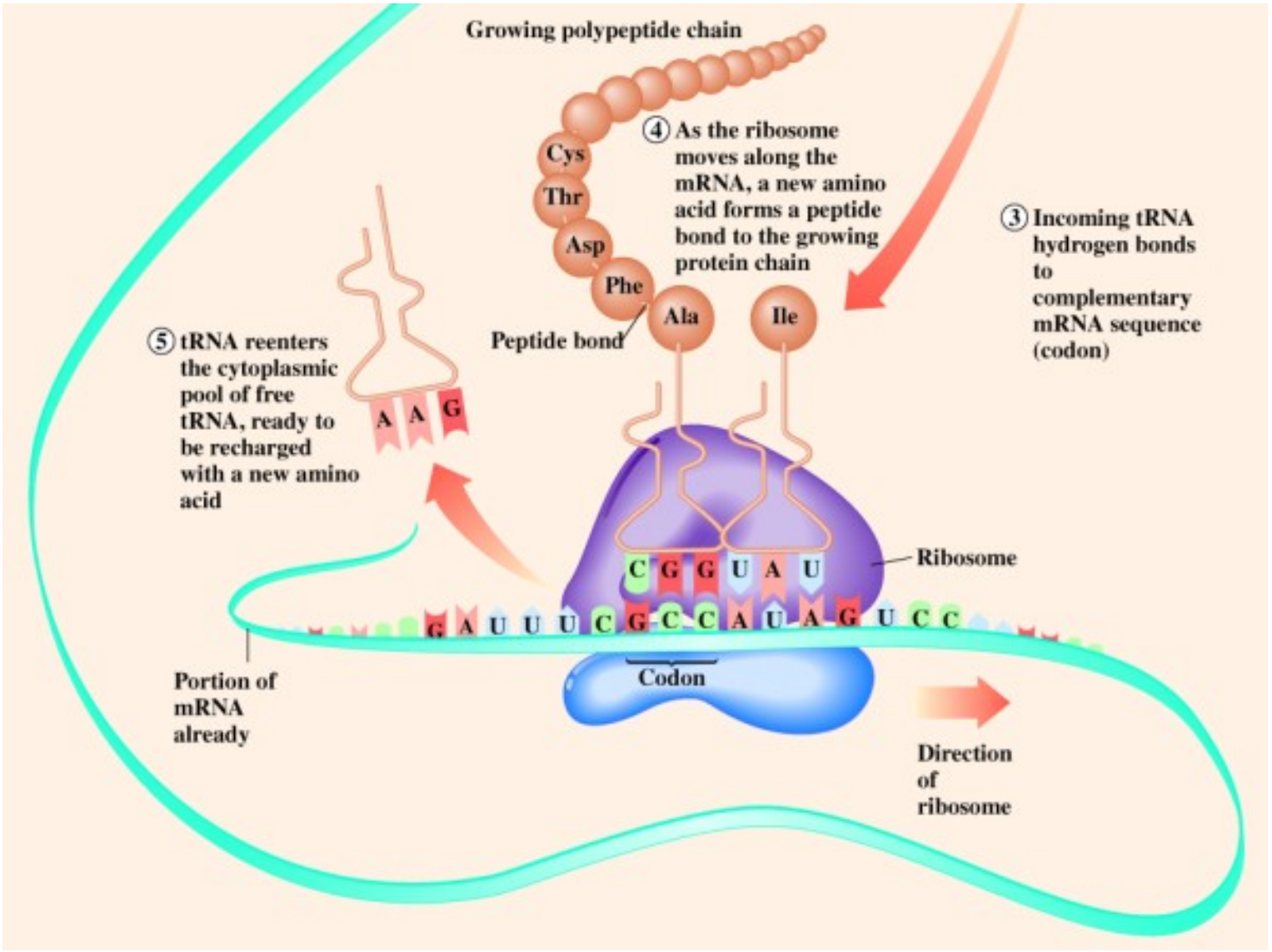
Overview: Translation



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Initiation of protein synthesis

Start codon on mRNA is usually AUG (Met)

Requires initiation factors (IF1, IF2 & IF3)

In bacteria further defined by:
Shine-Dalgarno sequence which base-pairs with ribosomal RNA

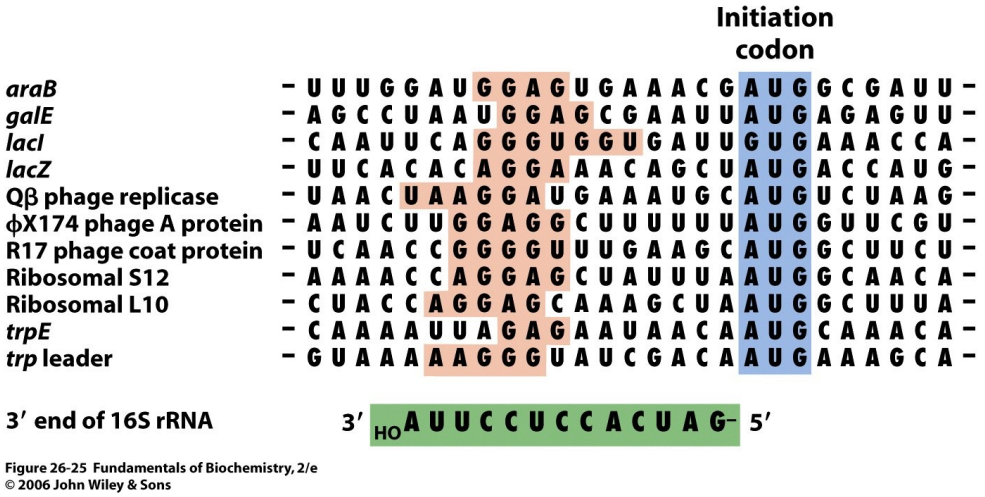

Figure 26-25 Fundamentals of Biochemistry, 2/e
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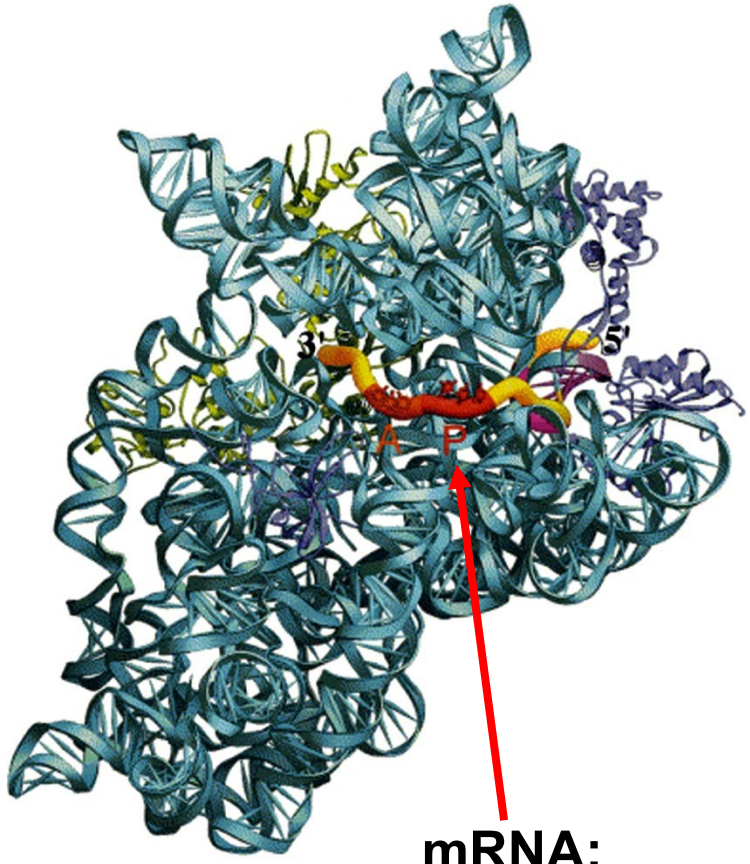


Figure 26-26 Fundamentals of Biochemistry, 2/e

mRNA: Start codon in P site

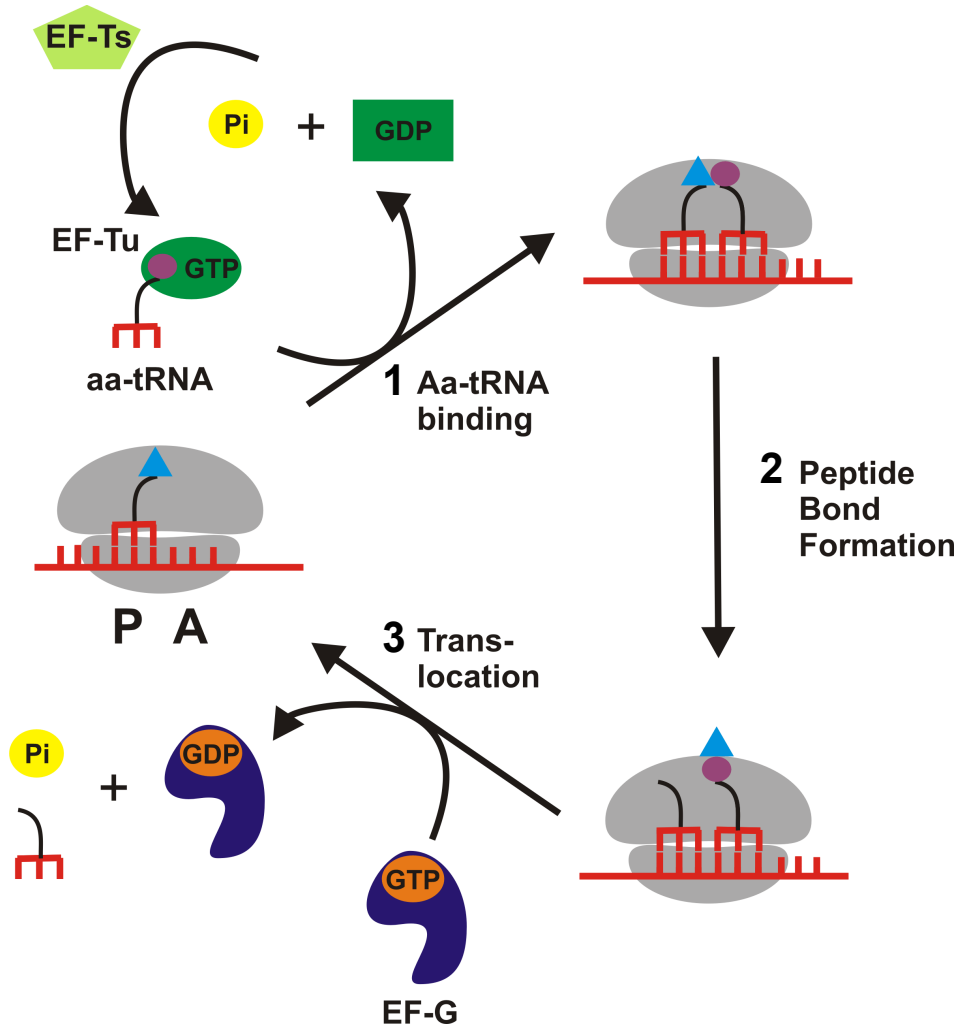
Elongation cycle

1. **Elongation factor Tu (EF-Tu)** delivers aminoacyl-tRNA to the A-site

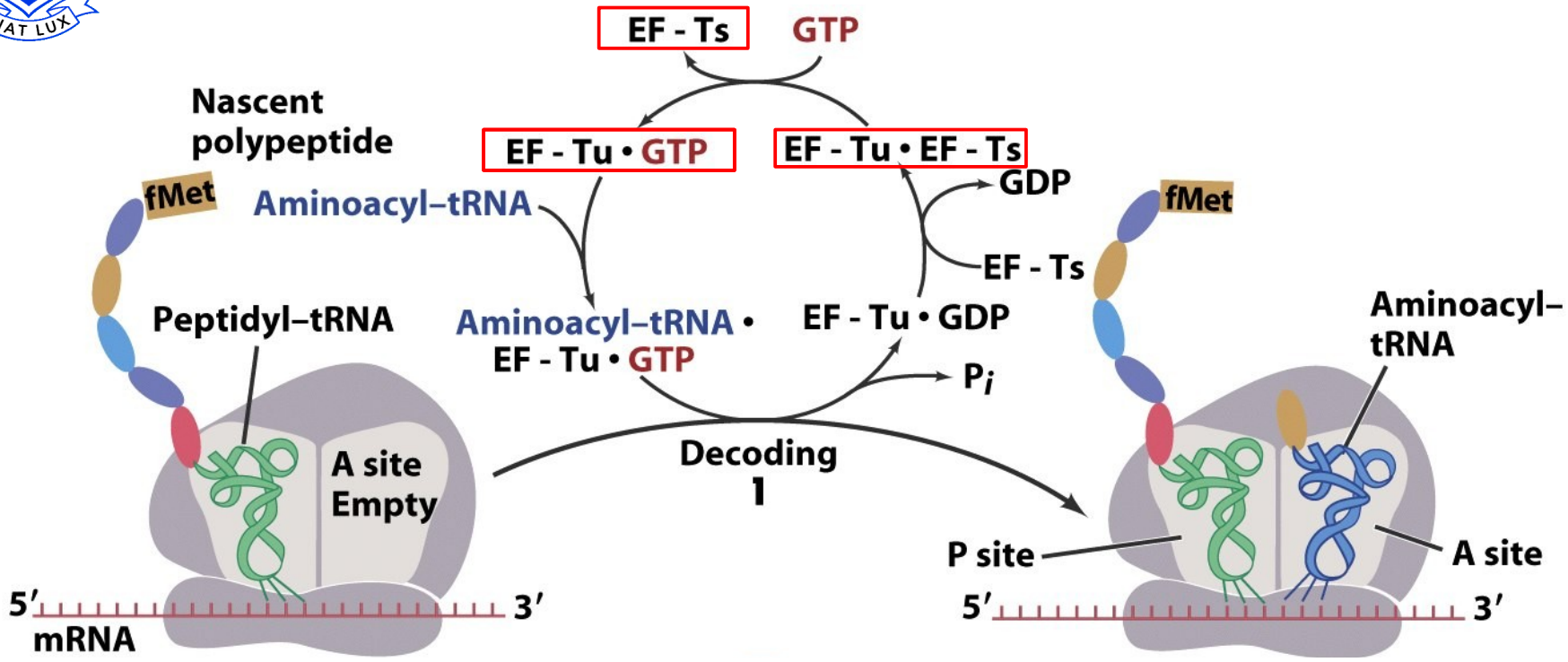
2. **Peptide bond** is formed between growing peptide chain and new amino acid

3. **Elongation factor G (EF-G)** catalyzes movement of the ribosome along mRNA (translocation)

⇒ Cycle can resume



Elongation factor Tu



EF-Tu as a G protein switch:

- EF-Tu-GTP binds aminoacyl-tRNA and brings it to the ribosome
- correct codon recognition triggers GTP hydrolysis by EF-Tu
- EF-Tu-GDP is recycled to EF-Tu-GTP by elongation factor Ts (EF-Ts)

Peptide Bond Formation

1. Amino group of aminoacyl-tRNA (A site) attacks ester bond between growing peptide chain and P-site tRNA

2. Peptide chain is transferred from P-site tRNA and to A-site tRNA (forms peptide bond)

Catalyzed by ribosomal RNA, i.e. ribozyme

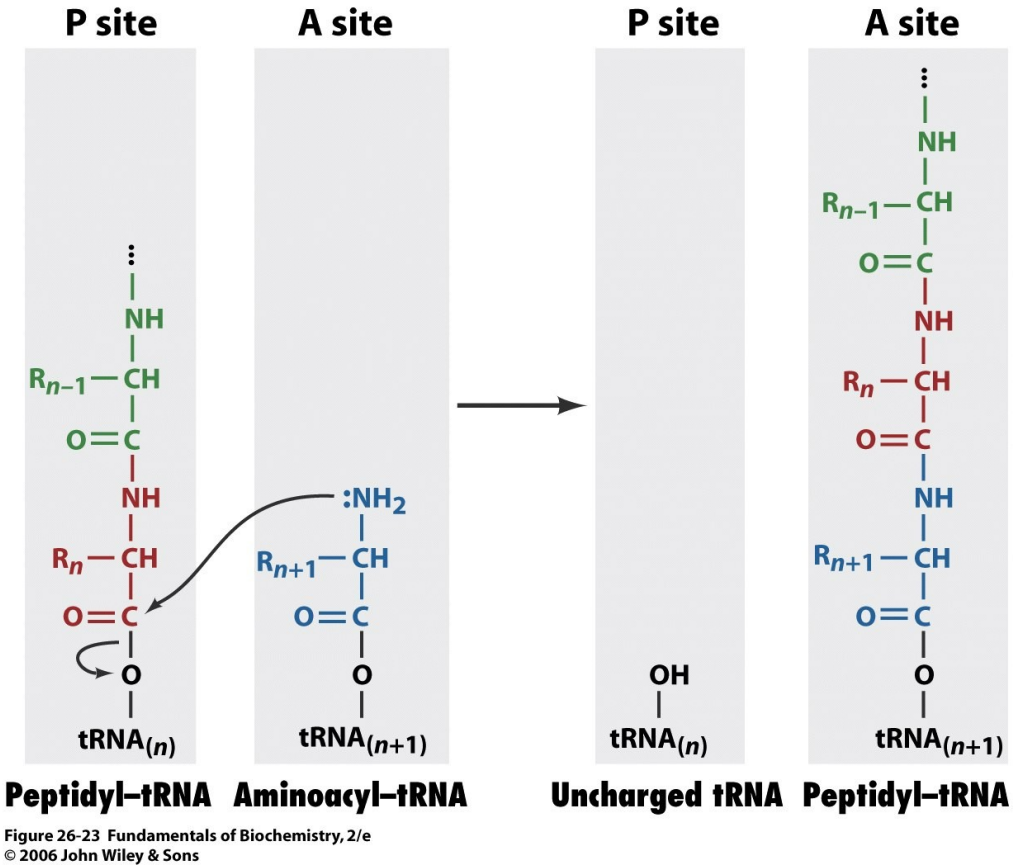


Figure 26-23 Fundamentals of Biochemistry, 2/e

⇒new peptidyl-tRNA is in A site

⇒deacylated (uncharged) tRNA in P site

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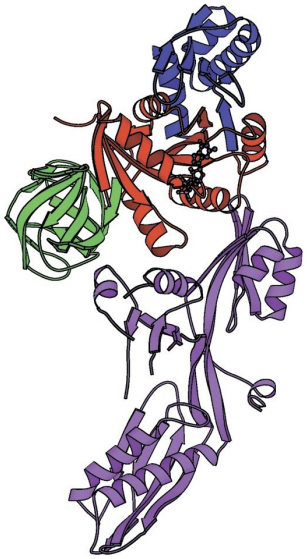
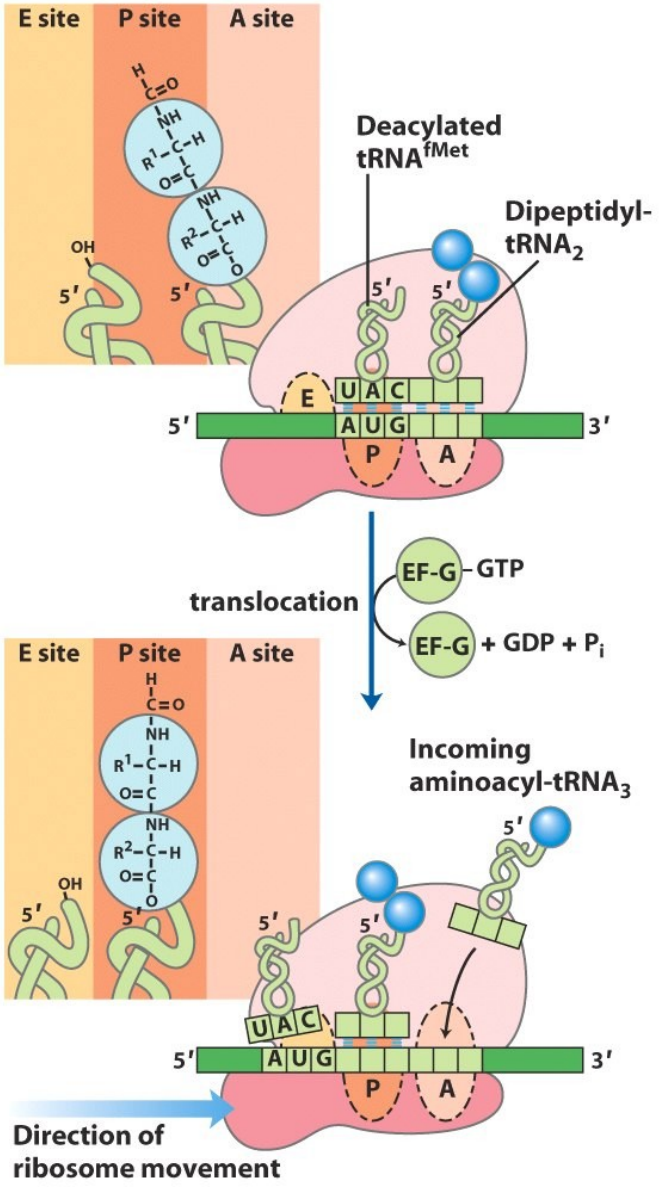


Translocation



Elongation factor G (EF-G) catalyzes

Simultaneous movement of:

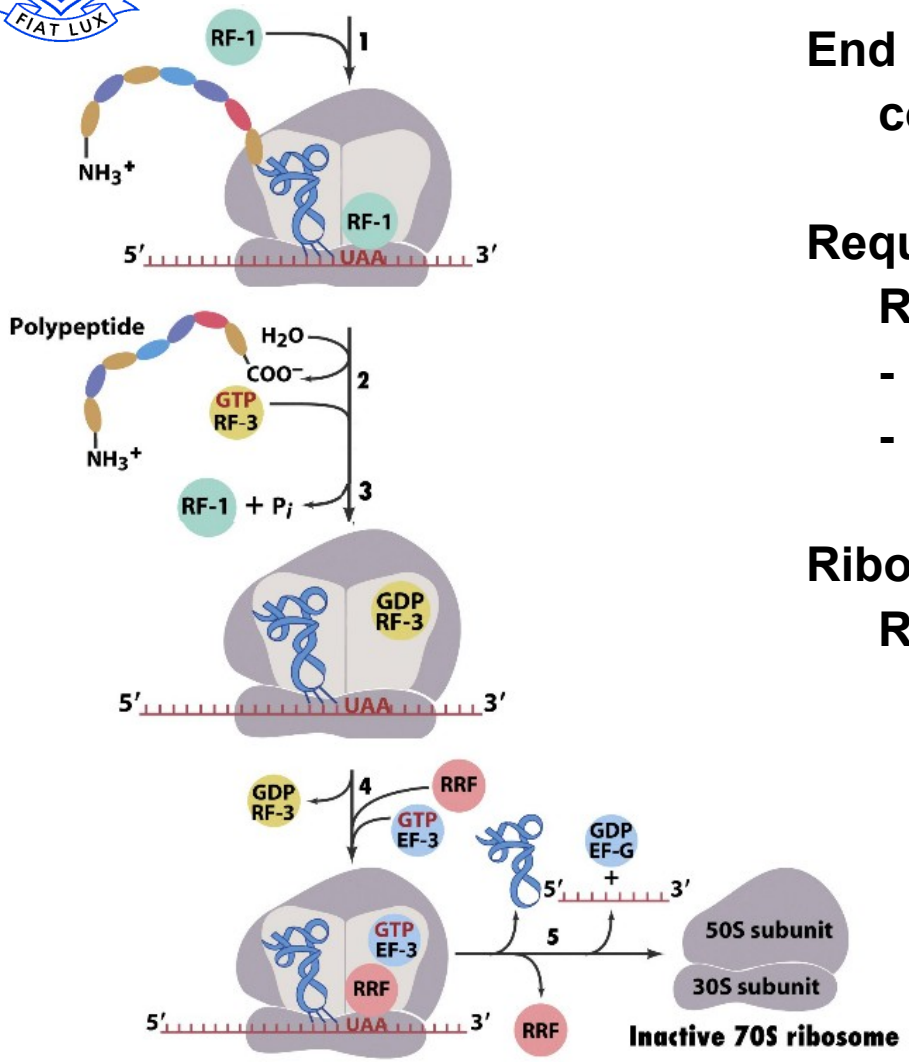
- **deacylated tRNA from P to E site (then dissociates from ribosome)**
- **peptidyl-tRNA from A to P site**
- **mRNA bound to the tRNAs**

EF-G also hydrolyzes GTP

⇒peptidyl-tRNA in the P site

⇒empty A site presenting next codon

Translation Termination



End of peptide chain indicated by stop codon in the A site

Requires release factors (RF1, RF2, RF3, RRF) and EF-G

- RF1/RF2 bind stop codon
- RF3 catalyzes RF1/RF2 removal

Ribosome Recycling:

RRF/EF-G catalyzes removal of tRNA and mRNA

RF3 and EF-G are GTPases