7.3.A1: tRNA-activating enzymes illustrate enzyme-substrate specificity and the role of phosphorylation.

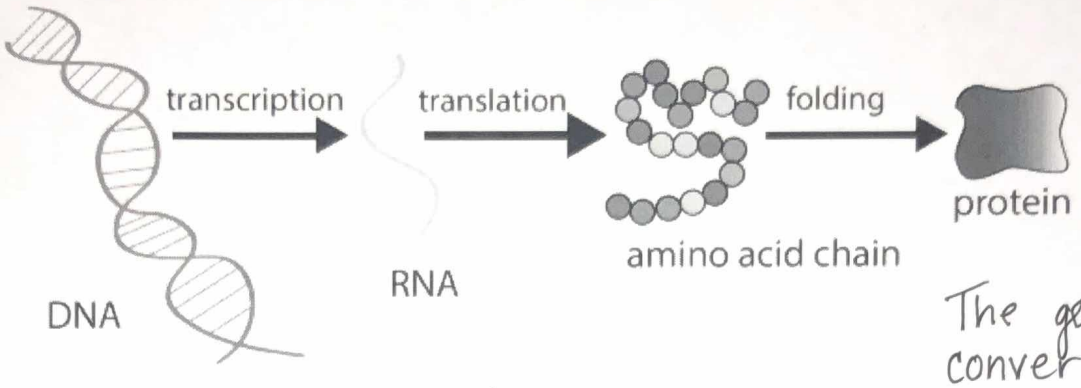
- State the role of the tRNA activating enzymes.
- Outline the process of attaching an amino acid to tRNA by the tRNA activating enzyme.

7.3.U3: Disassembly of the components follows termination of translation .

- Outline the process of translation termination, including the role of the stop codon.

3.1.A1: The causes of sickle cell anemia, including a base substitution mutation, a change to the base sequence of mRNA transcribed from it and a change to the sequence of a polypeptide in hemoglobin36358.

- State the cause of sickle cell anemia, including the name of differences in the Hb alleles.
- State the difference in amino acid sequences in transcription of normal and mutated Hb mRNA.
- Outline the consequences of the Hb mutation on the impacted individual.



The genetic code converts mRNA codons into amino acid.

The Genetic Code

in translation, the nucleic acid sequence codes for amino acids

codon = group of three mRNA nucleotides

Codon 1 met
Codon 2 thr
Codon 3 glu
Codon 4 leu
Codon 5 arg
Codon 6 ser
Codon 7 stop

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

The genetic code is:

Universal	Degenerate
a shared characteristic of all life. Homologous character! i.e. GGG codon codes for amino acid "Gly" in plants, fungus, animals, protist and bacteria!	more than 1 codon codes for the same amino acid. i.e. CGU, CGC, CGA and CGG all code for amino acid "Arg"

the nitty gritty of ...

Translation

Translation is the process of building a "polypeptide" chain of amino acids, guided by the sequence of codons on the mRNA.

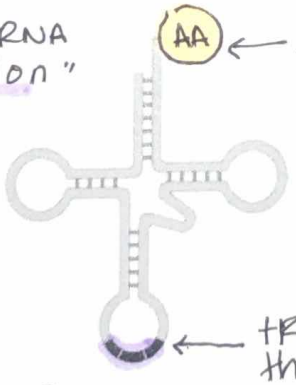
- In eukaryotic cells, the two main steps in protein synthesis occur in separate compartments: transcription in the nucleus and translation in the cytoplasm. mRNA moves out of the nucleus, to the cytoplasm, through pores in the nuclear membrane.
- In prokaryotic cells, there is no nucleus, and the chromosome is in direct contact with the cytoplasm, and protein synthesis can begin even while the DNA is still being transcribed.

Structures involved in translation:

- Messenger RNA (mRNA): carries the code from the DNA that will be translated into an amino acid chain

each set of 3 mRNA nucleotides is a "codon"

- Transfer RNA (tRNA) transports amino acids to the ribosome

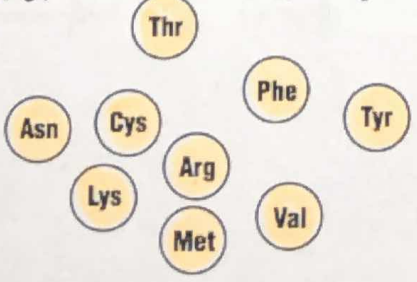


← the amino acid attaches to the tRNA here

← tRNA has a set of 3 nucleotides that are complementary to the mRNA codon... called "anticodon"

- Ribosomes globs made of rRNA and protein that will bind the mRNA and tRNA and attach amino acids together

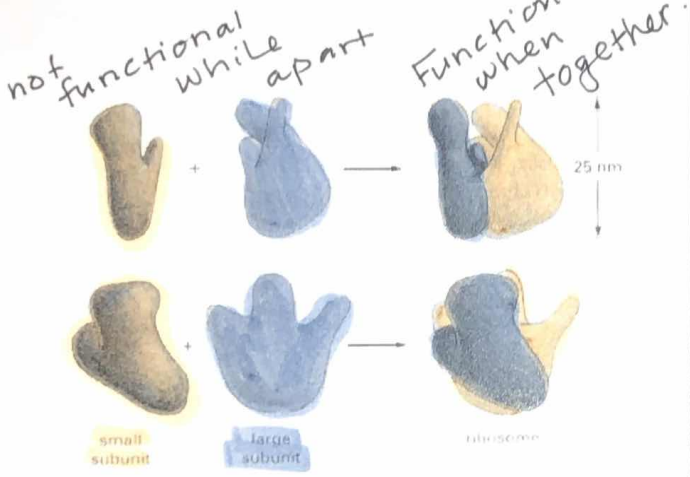
- Amino acids the monomer from which polypeptides are built.



there are 20 different amino acids used by living systems.

RIBOSOME Structure

- The ribosomal subunits are constructed of protein and ribosomal RNA (rRNA).
- Comprises two subunits in which there are grooves where the mRNA strand and polypeptide chain fit in.
- The subunits form a functional unit only when they attach to a mRNA molecule.



RIBOSOME Location

Ribosomes that synthesize proteins for use within the cell are FREELY suspended in the cytoplasm.

i.e. myosin
helicase
DNA polymerase
Ligase
actin

} all proteins used within the cell in which it is made

Ribosomes that synthesize proteins destined for:

- secretion (by exocytosis)
- the plasma membrane (e.g., cell surface receptors)
- lysosomes

are BOUND to the **endoplasmic reticulum**. As the polypeptide is synthesized, it is extruded into the lumen of the ER. Then, before these proteins reach their final destinations, they undergo a series of processing steps in the Golgi apparatus.

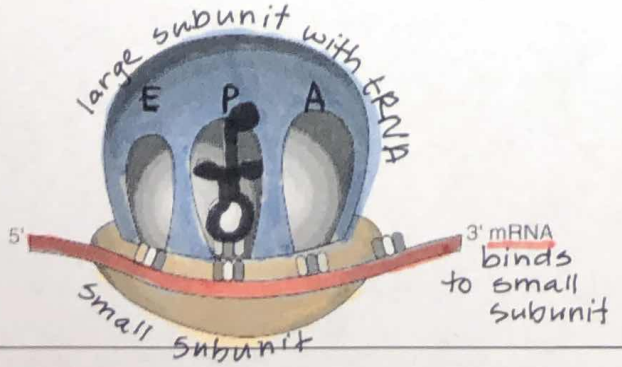
i.e. insulin — secreted
Na/k pump — in cell membrane

RIBOSOME Binding Sites

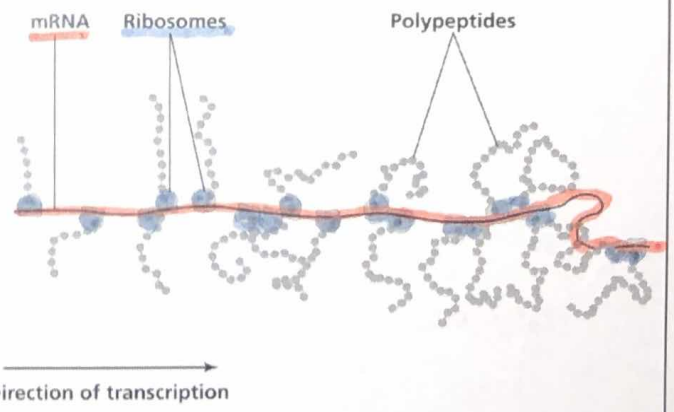
The mRNA attaches to the small ribosomal subunit in the "mRNA binding site"

The large ribosomal subunit has three tRNA binding sites:

- The A site
  tRNA anticodon complementary pairs with mRNA codon
- The P site
  ribosome forms a peptide bond between amino acids
- The E (exit) site
  tRNA leaves the ribosome



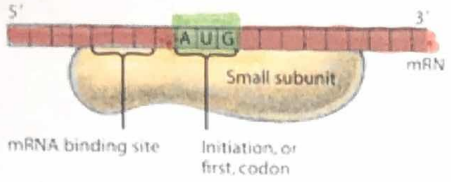
POLYSOME



multiple ribosomes all translating the same mRNA at the same time
∴ they are all making the same polypeptide

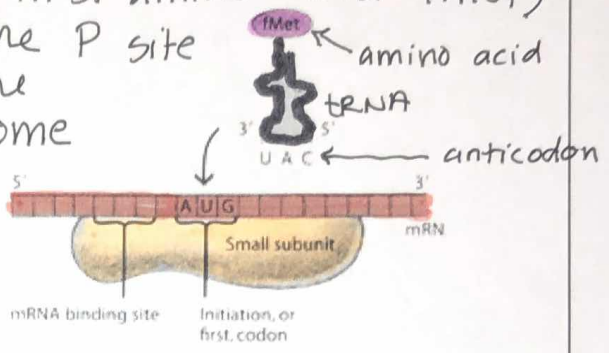
STEP 1: INITIATION

(1) the small ribosome subunit attaches to the mRNA at the "start codon," AUG.

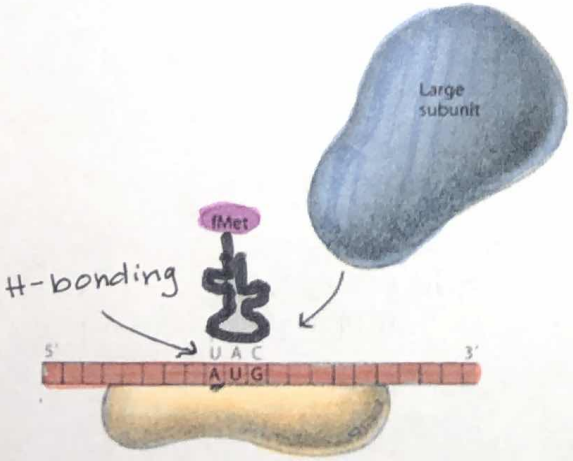


AUG is always the start. Anything before AUG is ignored.

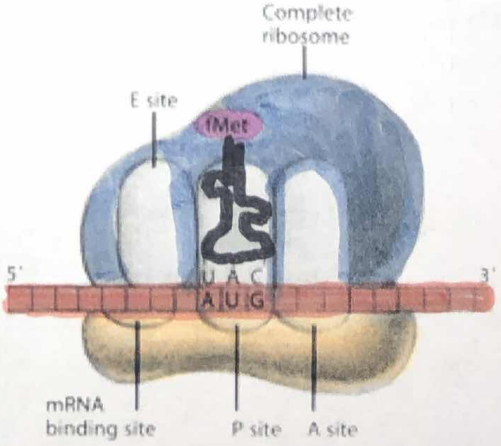
(2) a tRNA with an anticodon that is complementary to the AUG start codon brings the first amino acid (met) to the P site of the ribosome



(3) the large ribosomal subunit then binds



(4) the ribosome is built and ready for elongation

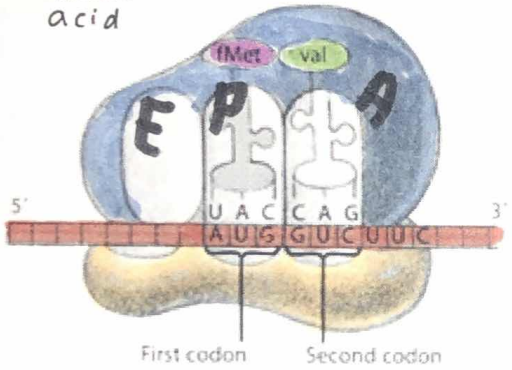


STEP 2: ELONGATION

1 Codon Recognition

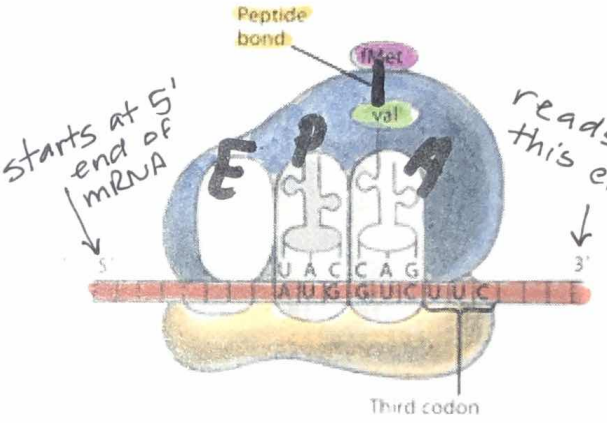
- mRNA codon in the A site
- tRNA with complementary ANTI-CODON hydrogen bonds to the codon in the A site
- the tRNA brings the appropriate amino acid to the ribosome

GUC codon = val amino acid



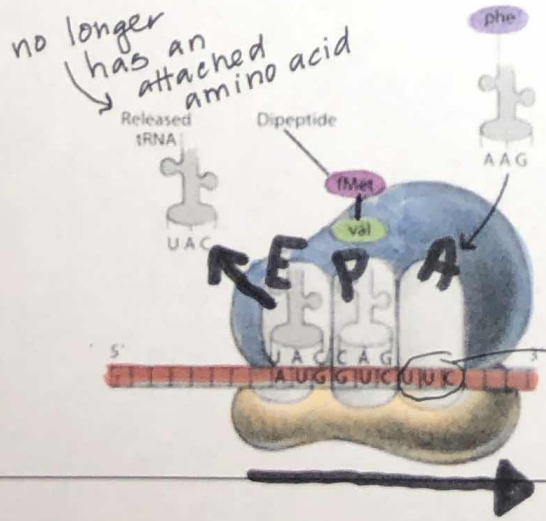
2 Bond formation

- The large ribosomal subunit makes a peptide bond (a type of covalent bond) between the amino acid in the A site and the amino acid in the P site.
- The mRNA molecule is read and new amino acids are added in the 5' → 3' direction



3 Translocation

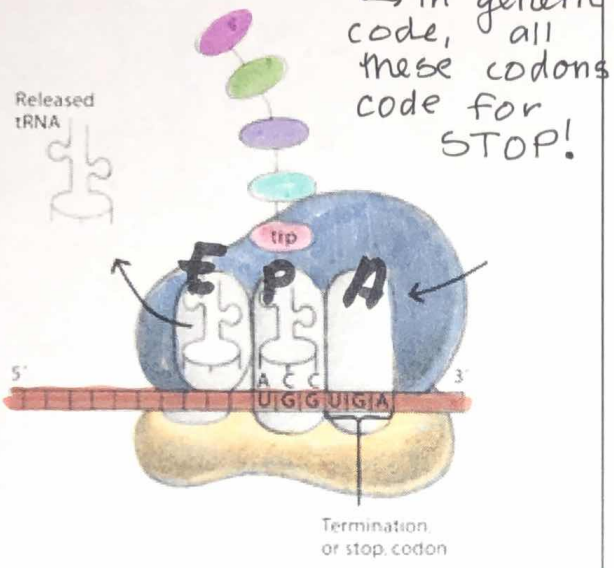
- The ribosome moves towards the 3' end of the mRNA strand
- Because of the move of the ribosome, there is a shift in the tRNA bindings site (A → P → E)
- tRNA leaves the ribosome from the E site



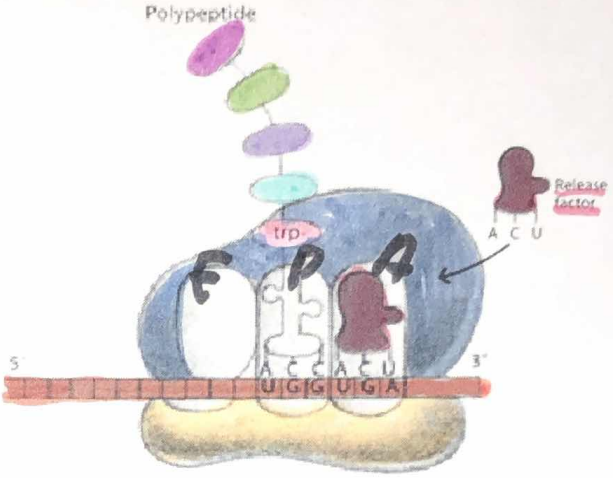
Repeat...
Repeat...
Repeat...
Repeat...
Repeat...
Repeat...
Repeat...

STEP 3: TERMINATION

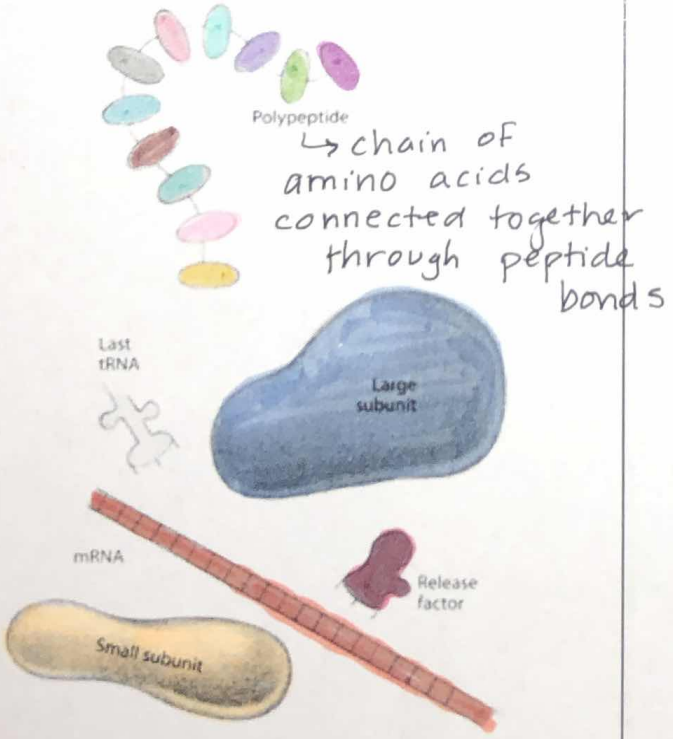
- Adding of amino acids to the polypeptide chain continues until there is a STOP codon on the RNA (UAA, UGA, or UAG)
  - ↳ in genetic code, all these codons code for STOP!

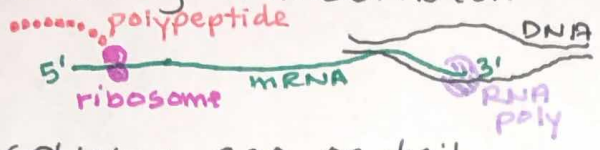


- A "release factor" binds to the stop codon and releases the amino acid chain.



- The ribosome dissociates into the two subunits and the amino acid chain can now fold into its secondary, tertiary and/or quaternary structure to become a functional protein



PROKARYOTE TRANSLATION	EUKARYOTE TRANSLATION
• no nucleus ∴ transcription and translation are not separated in time or space. mRNA being translated while still being transcribed.  • no splicing, cap or tail	• nucleus separates transcription and translation in time and space Transcription 1st — nucleus Translation 2nd — cytoplasm and/or rER • mRNA is spliced, cap & tail

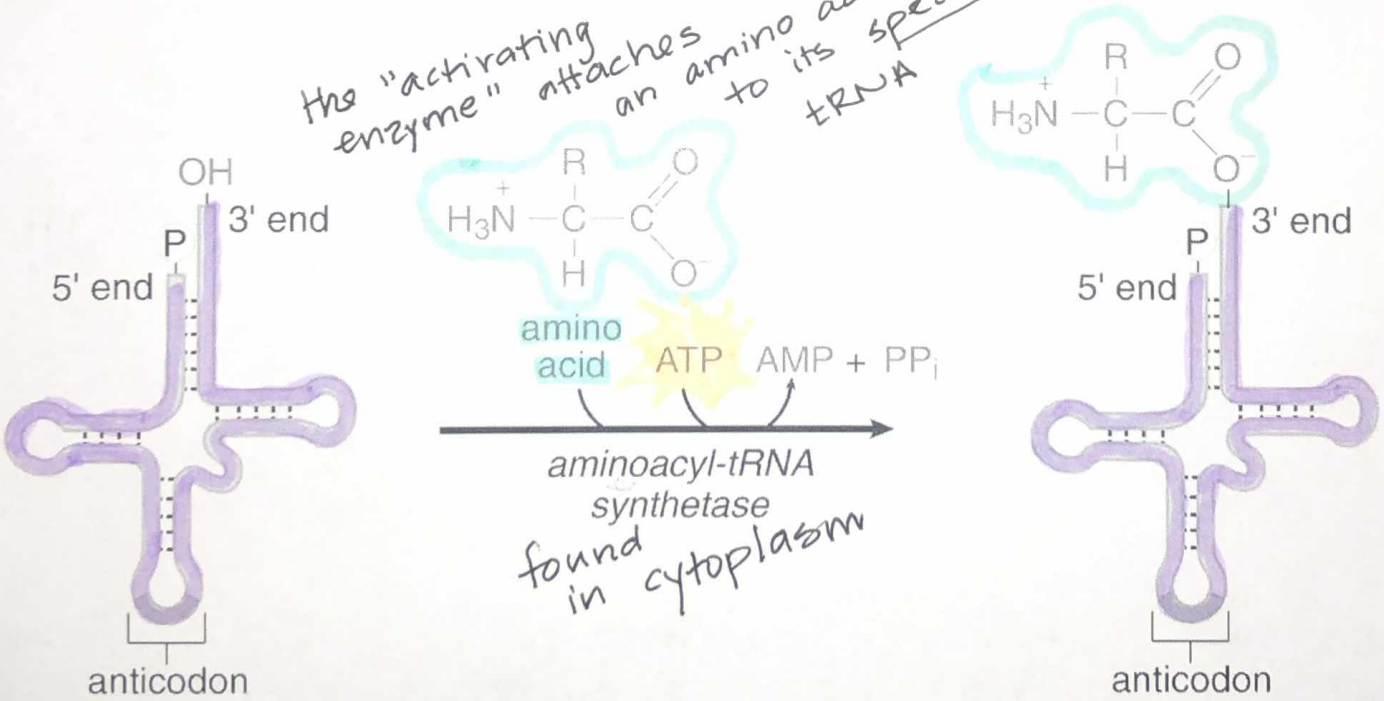
How does the tRNA "know" which amino acid to bring to the ribosome?

Each amino acid is joined to the correct tRNA by an enzyme called aminoacyl-tRNA synthetase (AKA tRNA activating enzyme")

There are 20 types of aminoacyl-tRNA synthetase enzymes in the cell – one for each type of amino acid

The enzyme requires ATP energy to attach the amino acid to the tRNA

the "activating enzyme" attaches an amino acid to its specific tRNA



"inactive tRNA" does not have an amino acid attached

"active tRNA" has amino acid attached