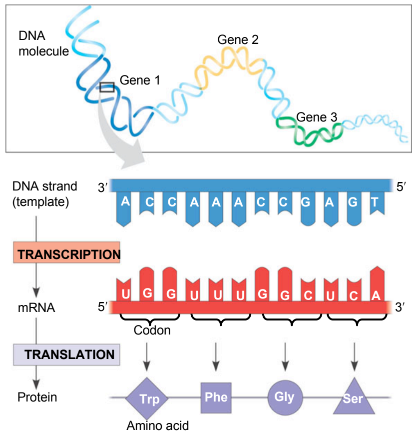
From Gene to Phenotype- part 2



Lecture Outline 11/7/05

- The central dogma:
  – DNA->RNA->protein
- Various types of RNA
- The genetic code
- Mechanisms of translation
  – Initiation, Elongation,Termination

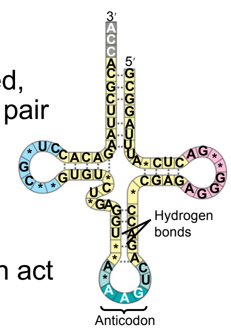
Exam 3 is next Monday. It will cover mitosis and meiosis, DNA synthesis, transcription, translation, genetics of viruses.
(chapters 12, 13, 16, 17, part of 18)

Four types of RNA

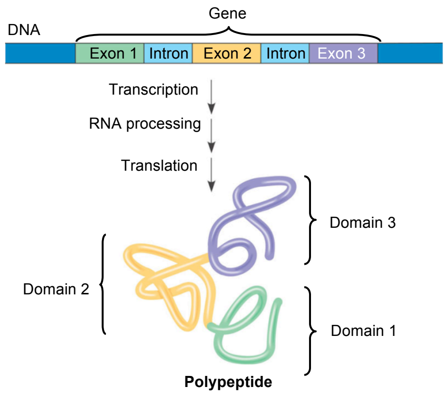
- mRNA
  – Messenger RNA, encodes the amino acid sequence of a polypeptide
- rRNA
  – Ribosomal RNA, forms complexes with protein called ribosomes, which translate mRNA to protein
- tRNA
  – Transfer RNA, transports amino acids to ribosomes during protein synthesis
- snRNA
  – Small nuclear RNA, forms complexes with proteins used in eukaryotic RNA processing

RNA will fold to specific shapes

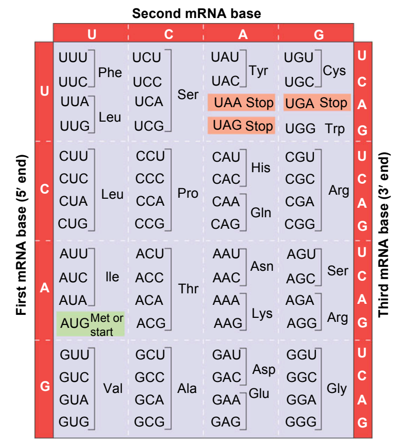
- Because RNA is single-stranded, parts of the molecule can base pair with other parts of the same molecule, causing it to fold into defined shapes.
- Some RNA molecules can even act as enzymes (ribozymes)



Correspondence between exons and protein domains



The genetic code



The code is a triplet code

- the new boy saw the big cat eat the hot dog

- The first evidence for triplet code came from deletion experiments
  - the new boy saw the big cat eat the hot dog
  - the neb oys awt heb igc ate att heh otd og

- Function could be restored by an insertion nearby
  - the new boy saw the big cat eat the hot dog
  - the neb oys aaw the big cat eat the hot dog

The code is a triplet code

- . . . or by two more deletions to get it back into the correct “reading frame”
  - the new boy saw the big cat eat the hot dog
  - the neb oys awt heb igc ate att heh otd og
  - the neb osa wte big cat eat the hot dog

Deciphering the Code

- Your book tells how people used synthetic mRNAs and *in vitro* translation to determine decipher the codons.
- E.g. UUUUUUUU -> phenylalanine only
  - UCUCUCUCUCU -> mix of leucine and serine
    - *Why is it a mixture?*

The code is redundant

- Several different codons encode the same amino acid

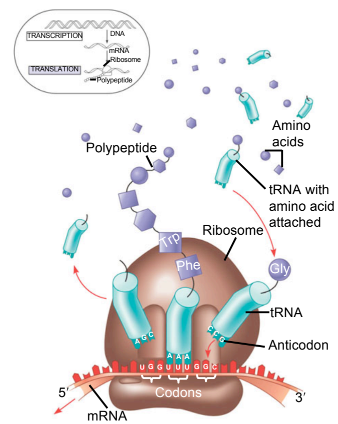
Second mRNA base

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

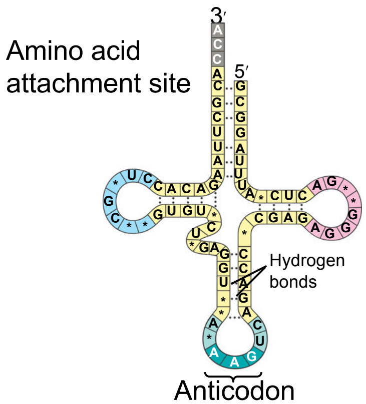
The code is comma free

- No punctuation between words.
  - Therefore deletions cause frameshifts
- It does have start and stop signals, however.
  - Start: AUG
  - Stop: UAG, UAA, UGA

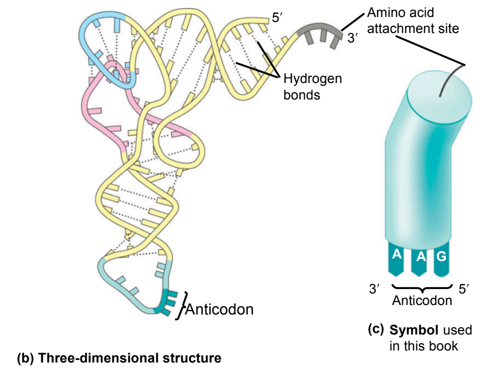
Translation: the basic concept



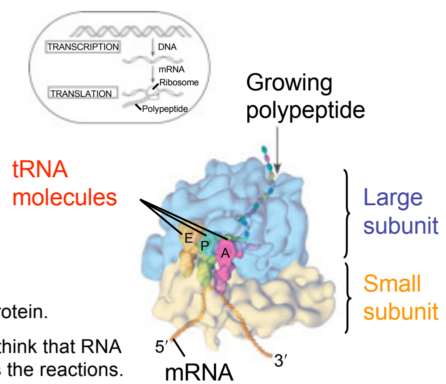
The structure of transfer RNA (tRNA)



tRNA



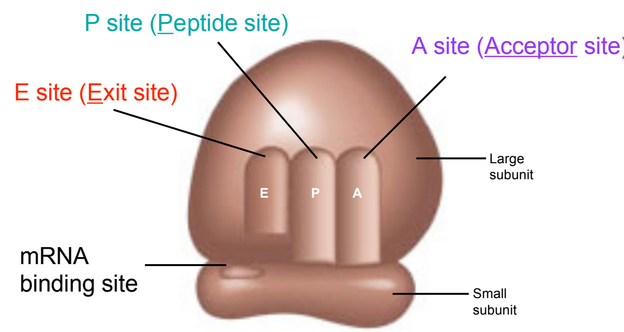
Ribosomes



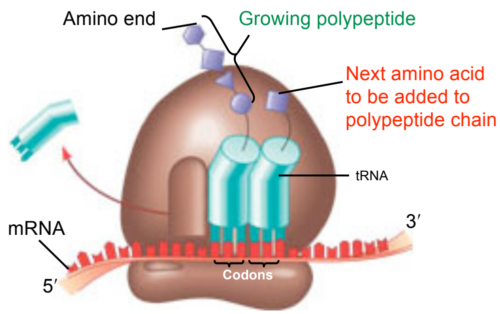
RNA + protein.

We now think that RNA catalyzes the reactions.

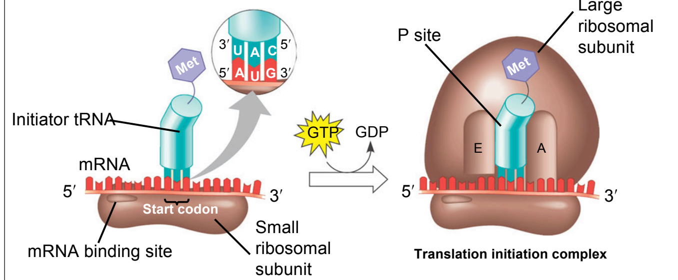
EPA model of a ribosome



New amino acids are added to the carboxyl end of the polypeptide



The initiation of translation

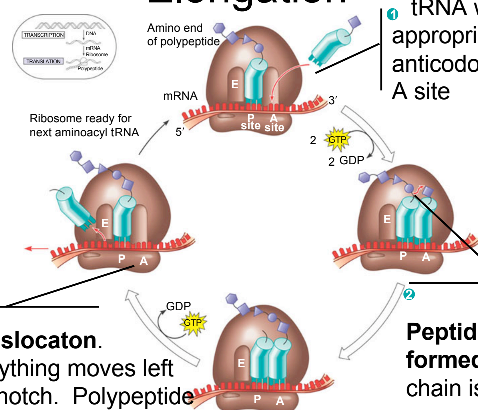


1. Small subunit binds just upstream of start

   Special initiator methionine binds to the start codon (AUG)

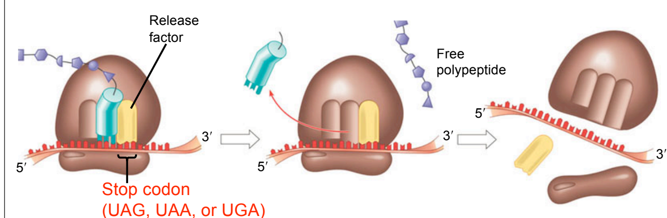
2. The large subunit can now bind to make the active ribosome

Elongation



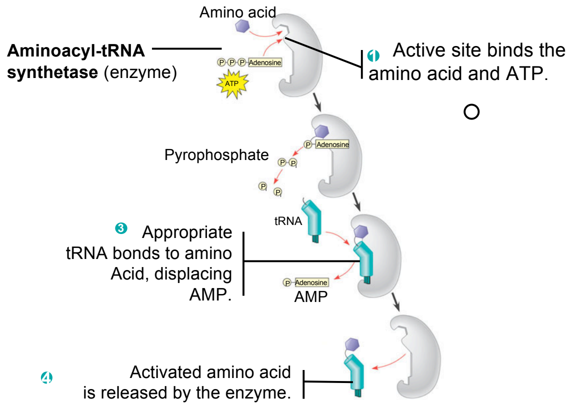
1. tRNA with appropriate anticodon binds at A site
2. **Peptide bond formed**. Growing chain is now at A
3. **Translocaton**. Everything moves left one notch. Polypeptide is now at P. Empty tRNA leaves the E site.

Termination



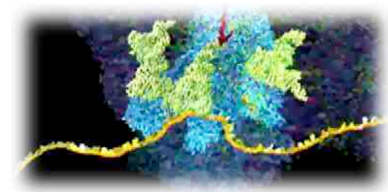
1. Stop codon has no matching tRNA
2. Release factor binds and separates the polypeptide from the tRNA
3. The ribosome separates from mRNA

An aminoacyl-tRNA synthetase joins a specific amino acid to a tRNA



See the Animation

- www.dnai.org



Inhibition of protein synthesis

Toxin	Mode of action	Target
Puromycin	forms peptidyl-puromycin, prevents translocation	Procaryotes
Tetracycline	blocks the A-site, prevents binding of aminoacyl tRNAs	Procaryotes
Chloramphenicol	blocks peptidyl transfer	Procaryotes
Cycloheximide	blocks peptidyl transferase	Eucaryotes
Streptomycin	inhibits initiation at high concentrations	Procaryotes
Diphtheria toxin	catalyzes ADP-ribosylation of residue in eEF2	Eucaryotes
Erythromycin	binds to 50S subunit, inhibits translocation	Procaryotes
Ricin	inactivates 60S subunit, depurinates an adenosine in 23S rRNA	Eucaryotes

NOTE: Prokaryotes (this generally includes protein synthesis in mitochondria and chloroplasts)

Summary of transcription and translation in a eukaryotic cell