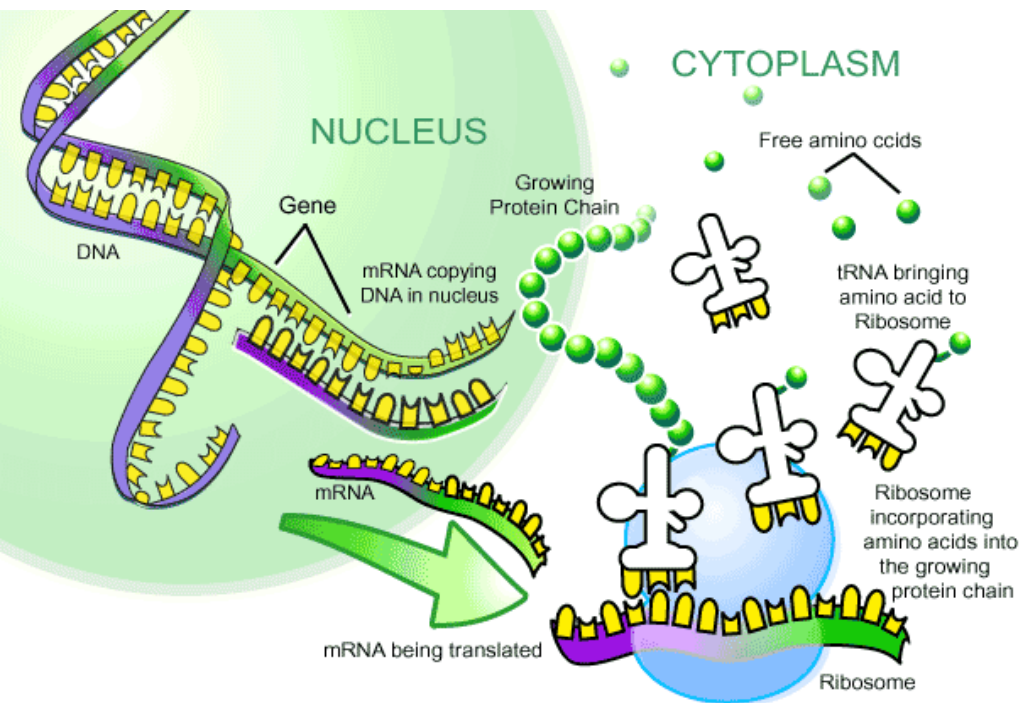
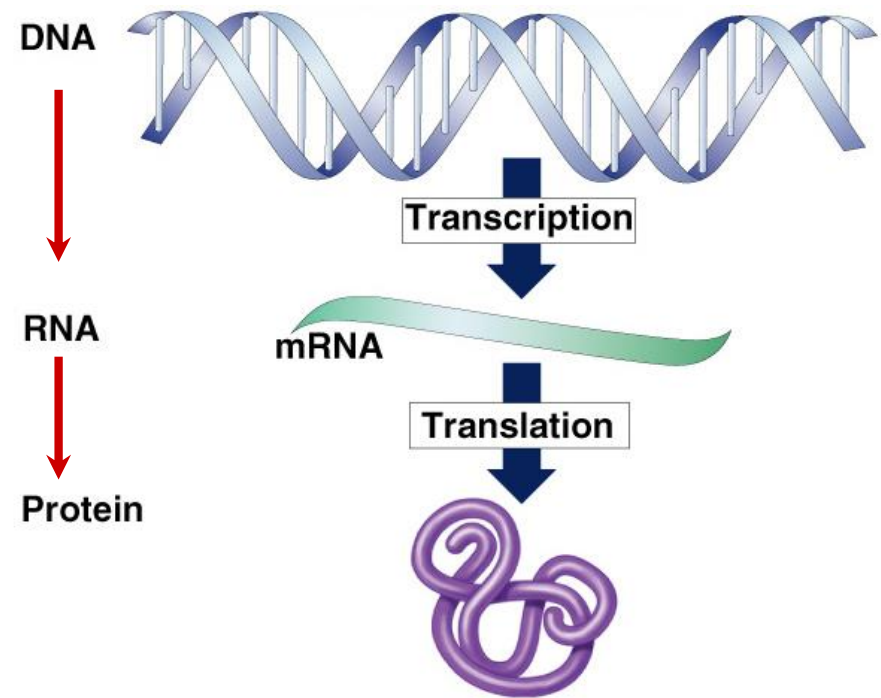


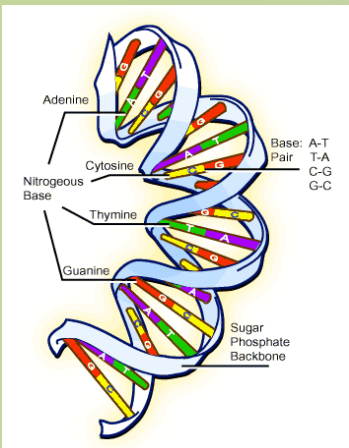
# RNA and Protein Synthesis



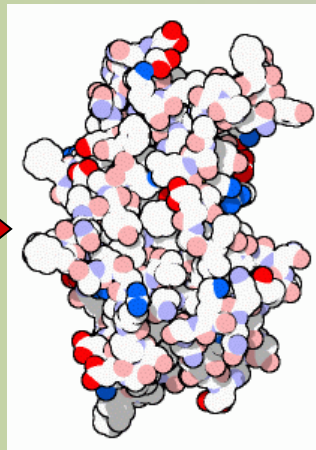
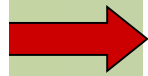
Ch. 13.1

# What do genes code for?

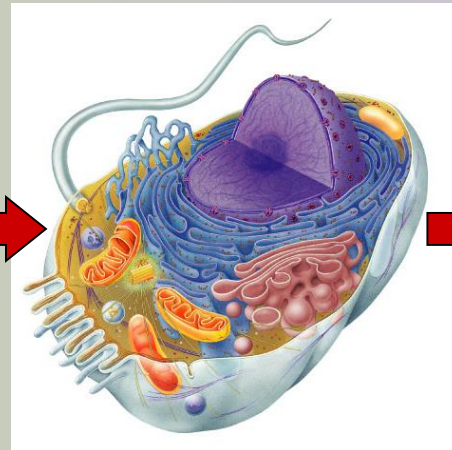
- **How does DNA code for cells & bodies?**
  - ◆ how are cells and bodies made from the instructions in DNA



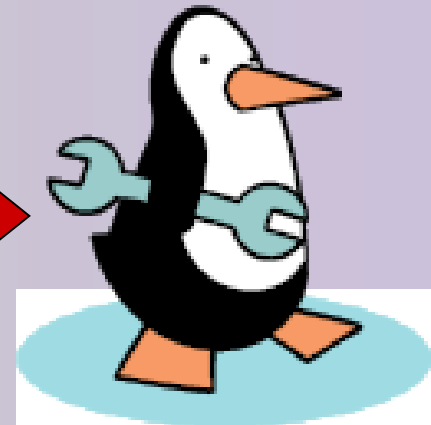
**DNA**



**proteins**

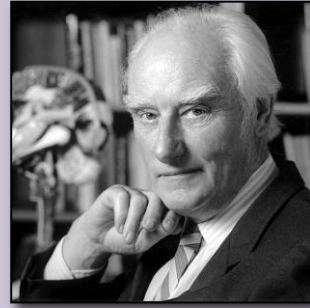


**cells**

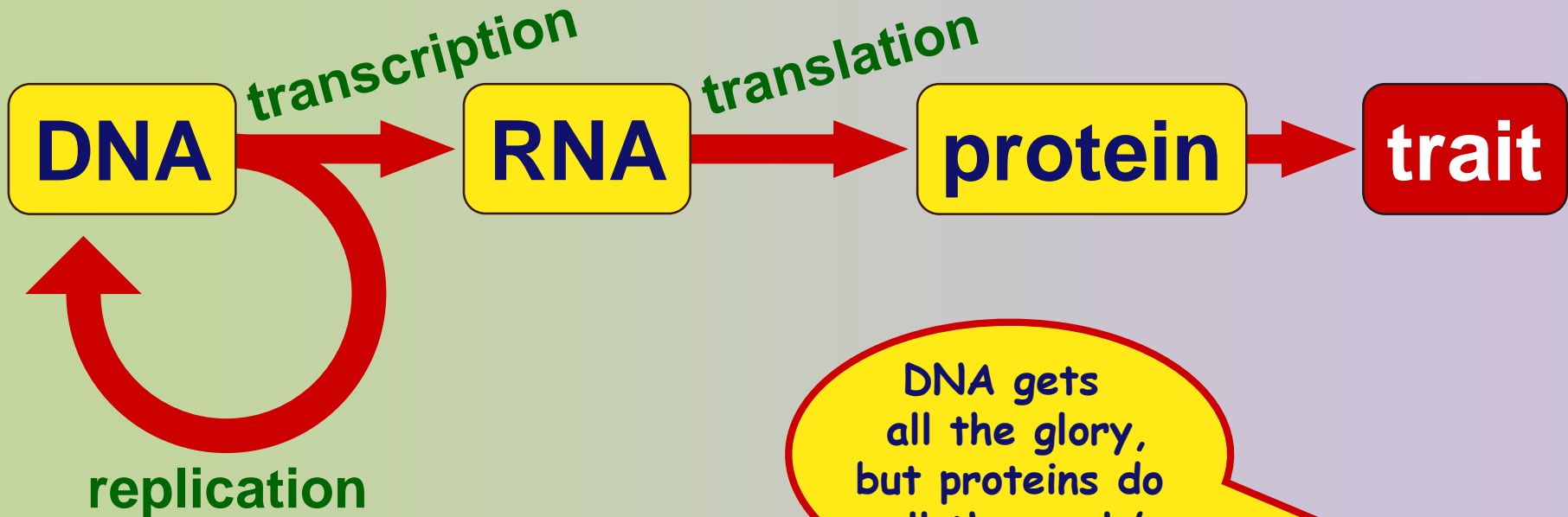


**bodies**

# The “Central Dogma”



- Flow of genetic information in a cell
  - How do we move information from DNA to proteins?



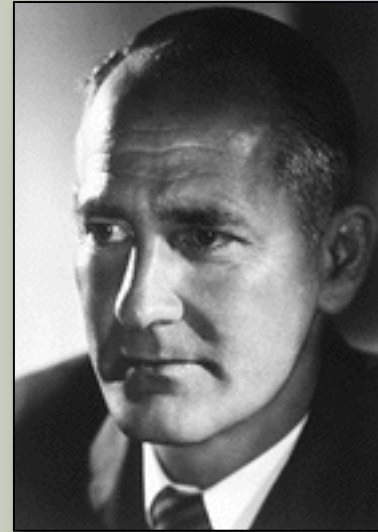
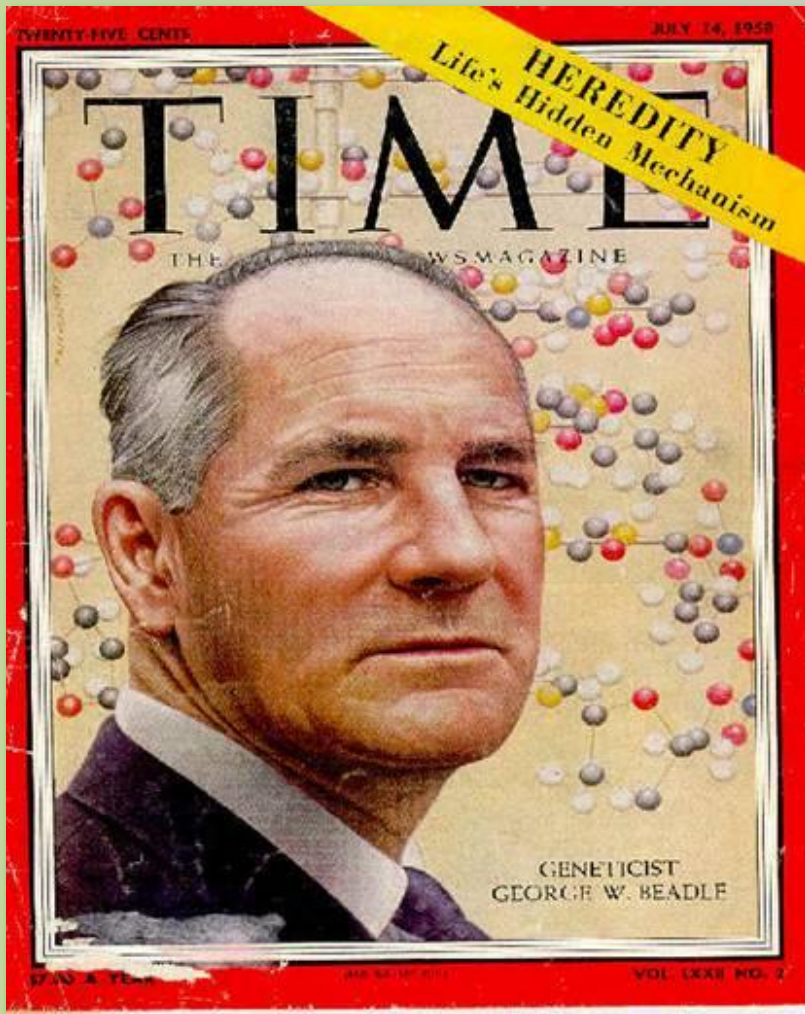
DNA gets  
all the glory,  
but proteins do  
all the work!



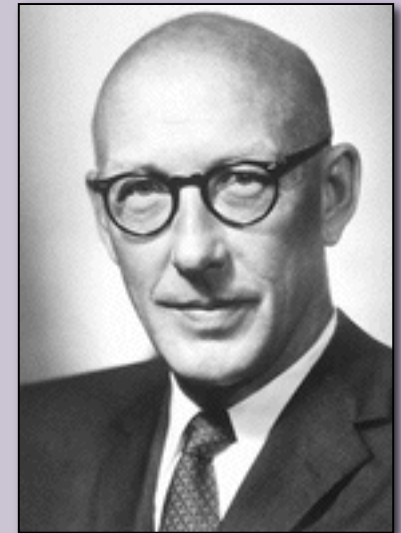
# Beadle & Tatum

1941 | 1958

one gene : one enzyme hypothesis



George Beadle



Edward Tatum

"for their discovery that genes act by regulating definite chemical events"

# Beadle & Tatum

Wild-type  
*Neurospora*

Minimal  
medium

asexual  
spores

X rays or ultraviolet light  
**create mutations**

Growth on  
complete  
**positive control**

spores

Select one of  
the spores

Test on minimal  
medium to confirm  
**negative control**

Grow on  
complete medium

Minimal media supplemented with...

**experimentals**

**amino acid  
supplements**

Pyridoxine  
benzoic acid

Choline  
Inositol

Nucleic  
acid  
Folic  
acid

Arginine  
Niacin

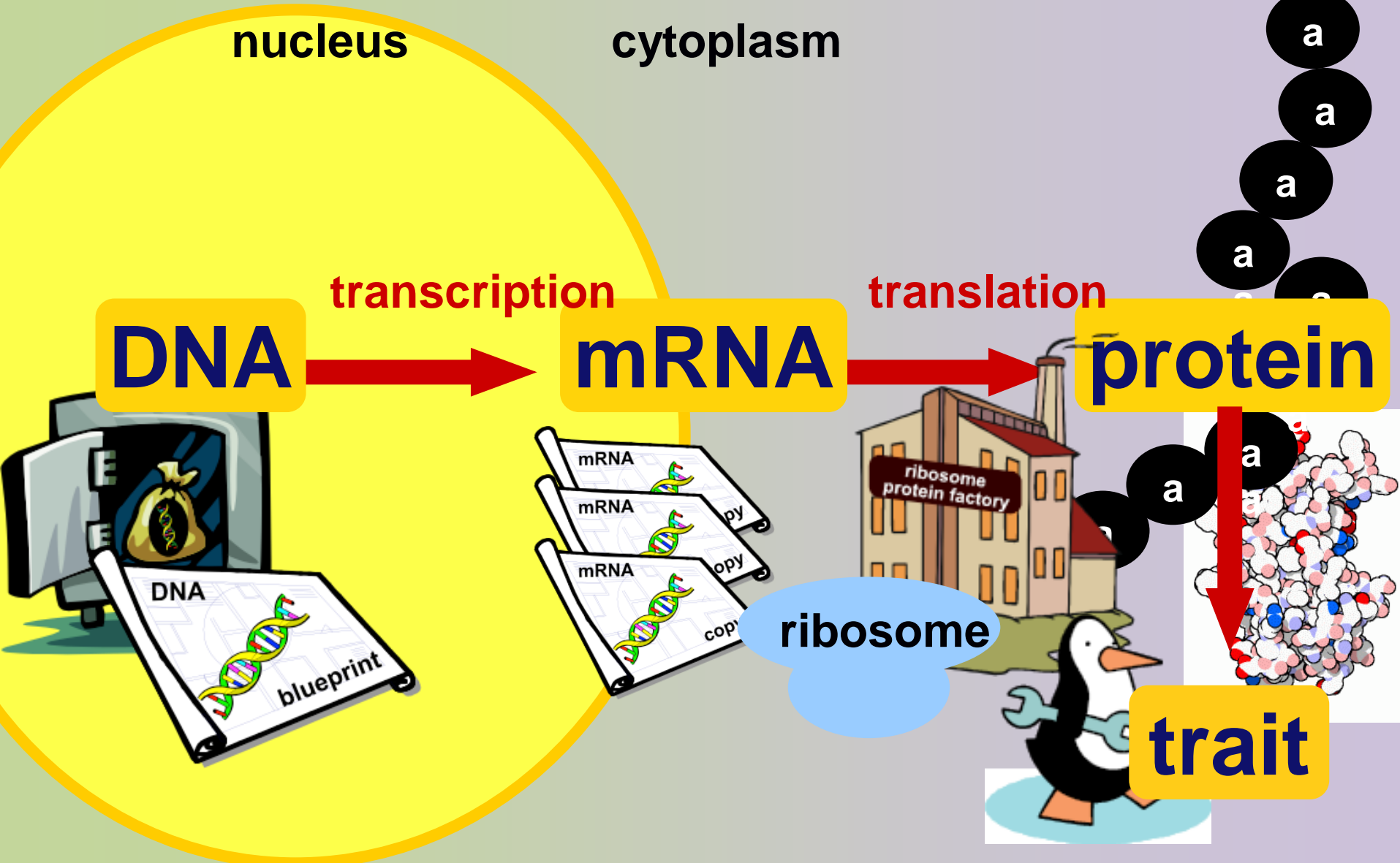
Riboflavin  
Thiamine

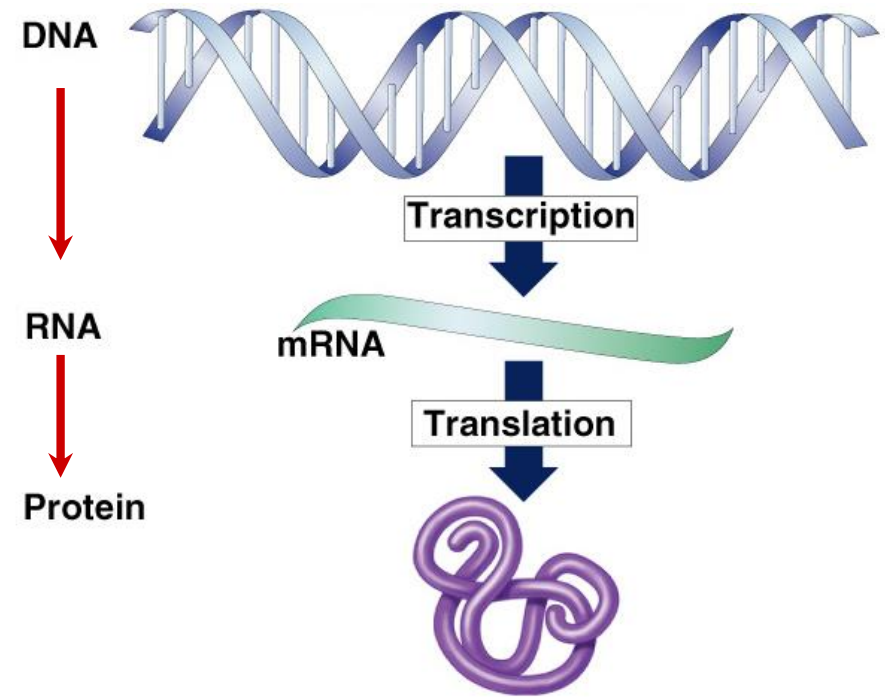
Minimal  
control

**mutation identified**



# From gene to protein





# Transcription

From DNA nucleic acid language  
to RNA nucleic acid language

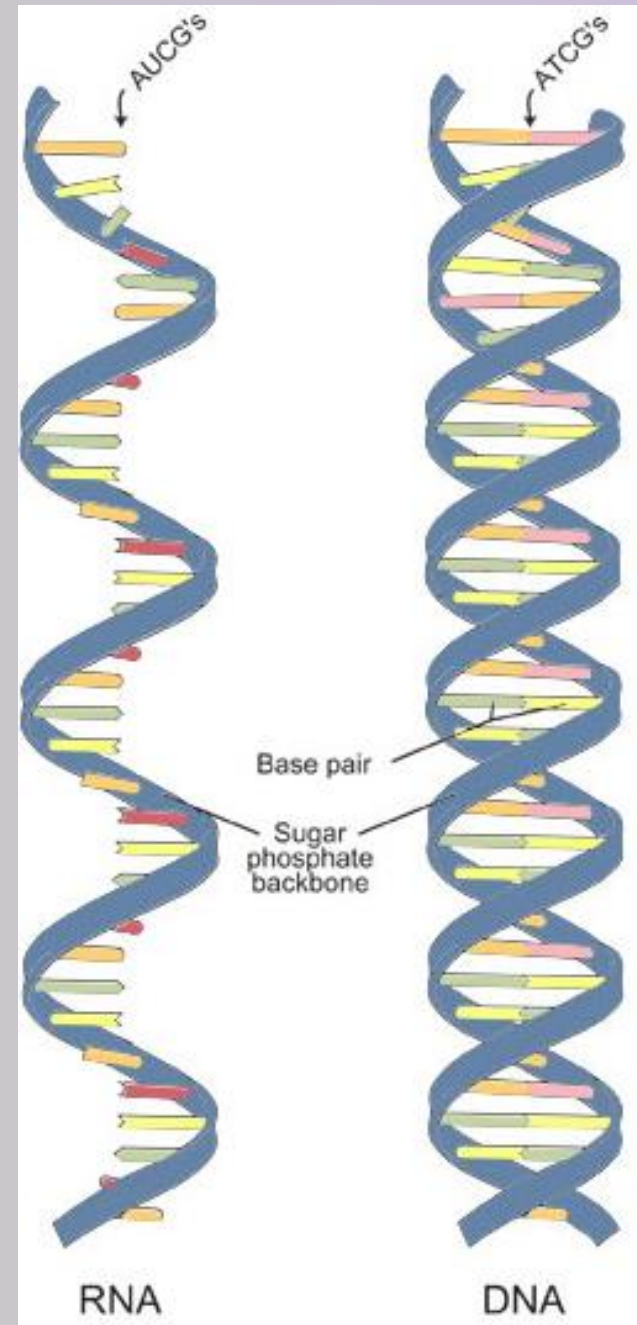
# RNA

- ribose sugar
- N-bases
  - uracil instead of thymine
  - U : A
  - C : G
- single stranded
- lots of RNAs
  - mRNA, tRNA, rRNA, siRNA...

**DNA**

transcription

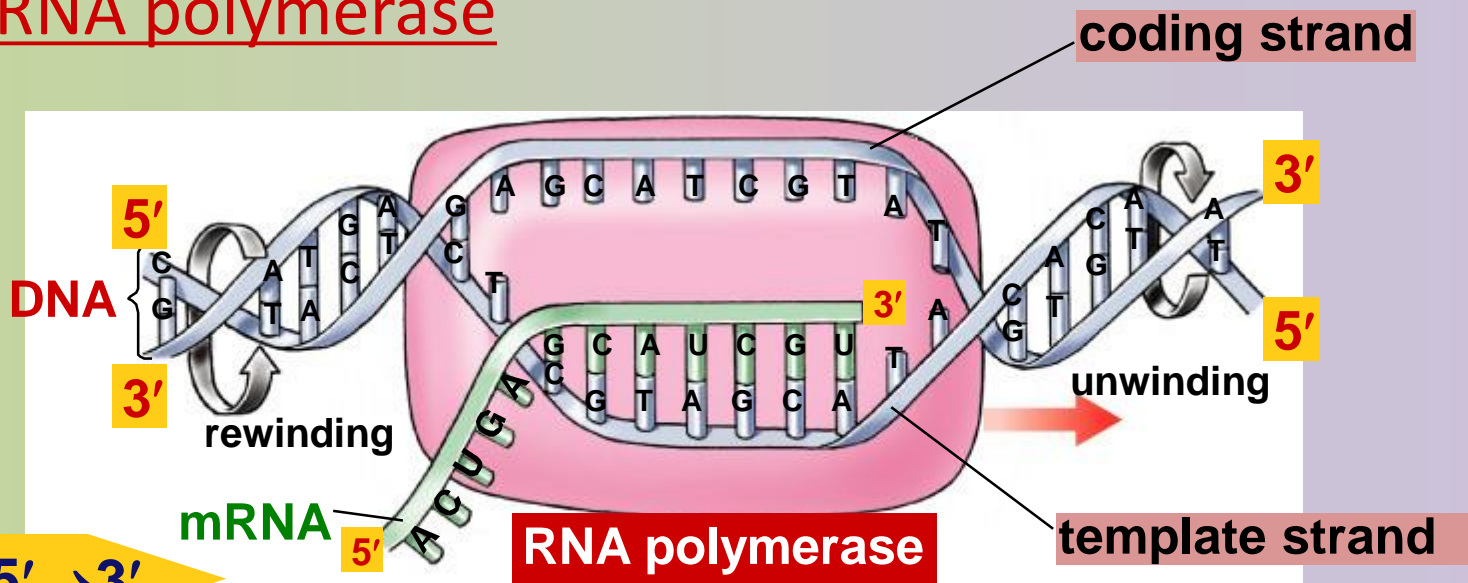
**RNA**





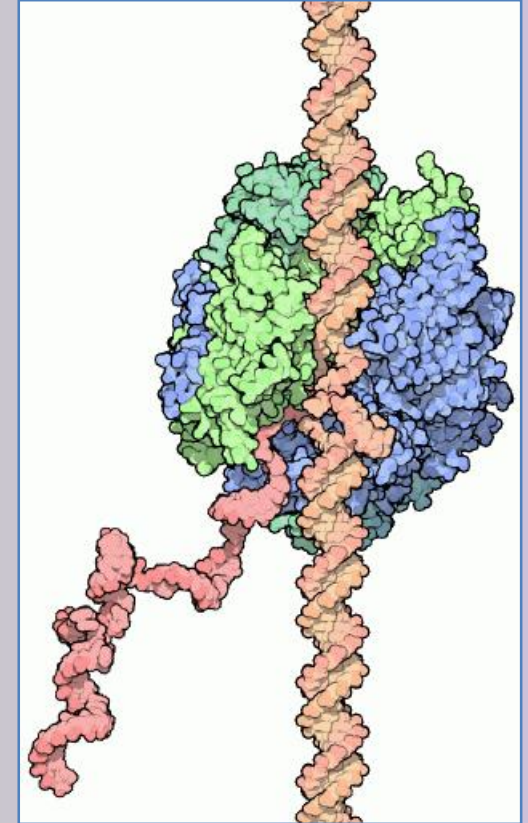
# Transcription

- Making mRNA
  - transcribed DNA strand = template strand
  - untranscribed DNA strand = coding strand
    - same sequence as RNA
  - synthesis of complementary RNA strand
  - enzyme
    - RNA polymerase



# RNA polymerases

- 3 RNA polymerase enzymes
  - RNA polymerase 1
    - only transcribes rRNA genes
    - makes ribosomes
  - RNA polymerase 2
    - transcribes genes into mRNA
  - RNA polymerase 3
    - only transcribes tRNA genes
  - each has a specific promoter sequence it recognizes



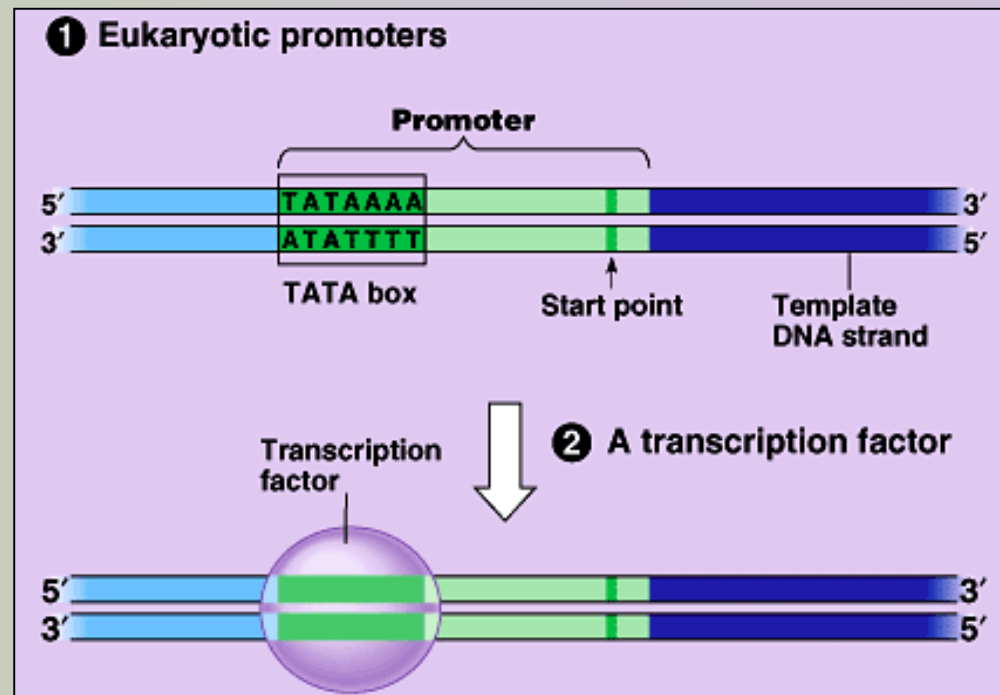
# Which gene is read?

- Promoter region

- binding site before beginning of gene
- TATA box binding site
- binding site for RNA polymerase & transcription factors

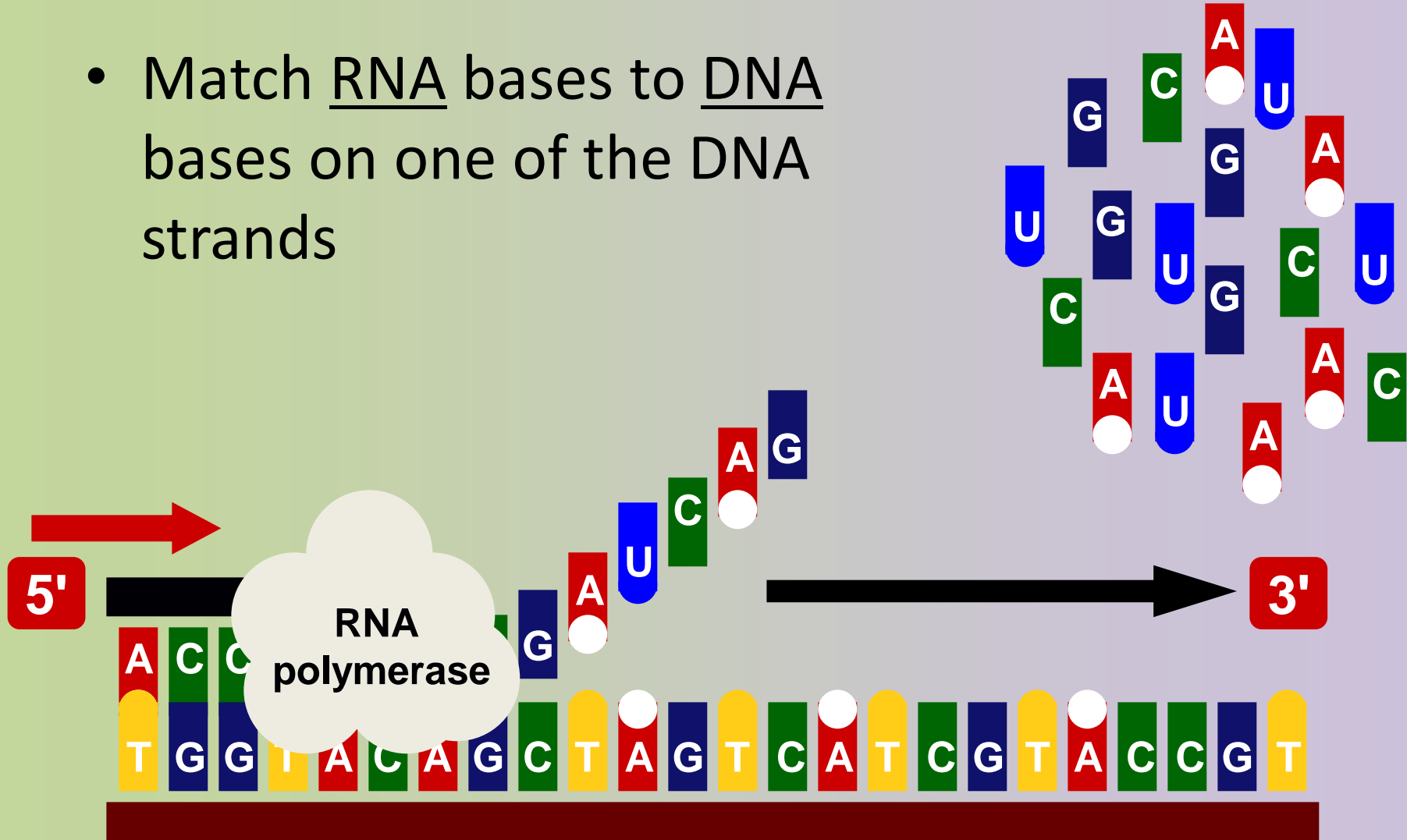
- Enhancer region

- binding site far upstream of gene
  - turns transcription on HIGH



# Matching bases of DNA & RNA

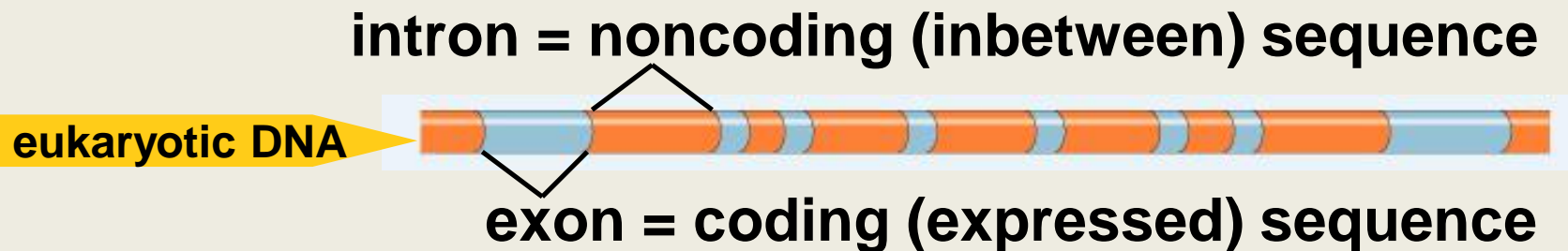
- Match RNA bases to DNA bases on one of the DNA strands



# Eukaryotic genes have junk!

- Eukaryotic genes are not continuous
  - exons = the real gene
    - expressed / coding DNA
  - introns = the junk
    - In between sequence

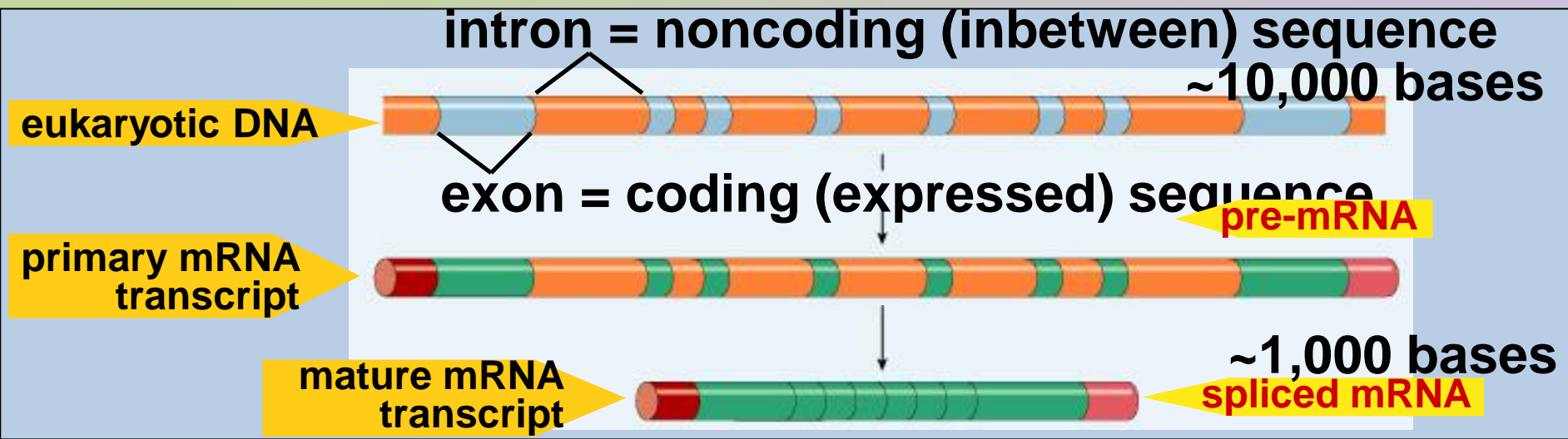
introns  
come out!





# mRNA splicing

- Post-transcriptional processing
  - eukaryotic mRNA needs work after transcription
  - primary transcript = pre-mRNA
  - mRNA splicing
    - edit out introns
  - make mature mRNA transcript



# Splicing must be accurate

- No room for mistakes!
  - a single base added or lost throws off the reading frame

AUGCGGCTATGGGUCCGAUAAGGGCCAU

AUGCGGUCCGAUAAGGGCCAU

AUG | CGG | UCC | GAU | AAG | GGC | CAU

Met | Arg | Ser | Asp | Lys | Gly | His

AUGCGGCTATGGGUCCGAUAAGGGCCAU

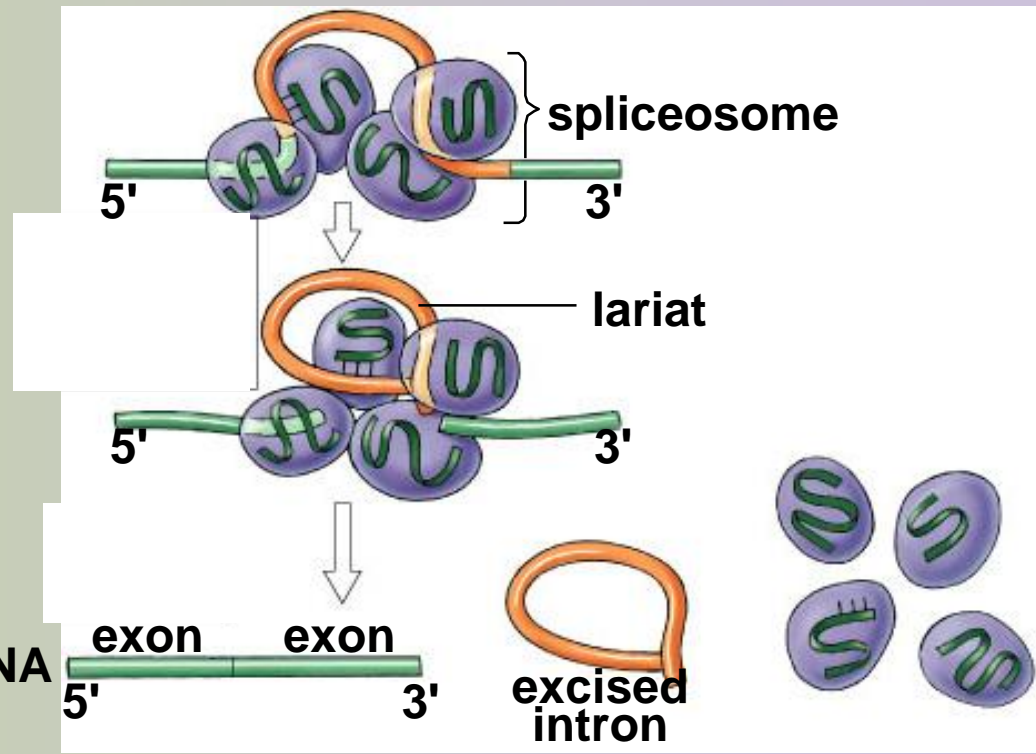
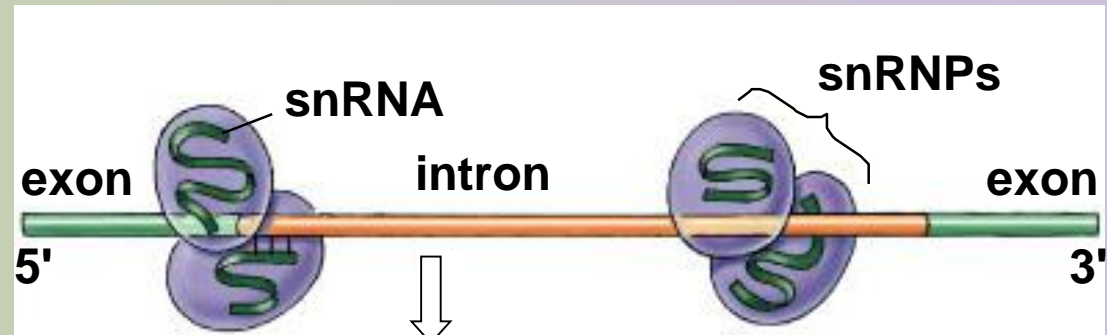
AUGCGGUCCGAUAAGGGCCAU

AUG | CGG | GUC | CGA | UAA | GGG | CCA | U

Met | Arg | Val | Arg | STOP |

# RNA splicing enzymes

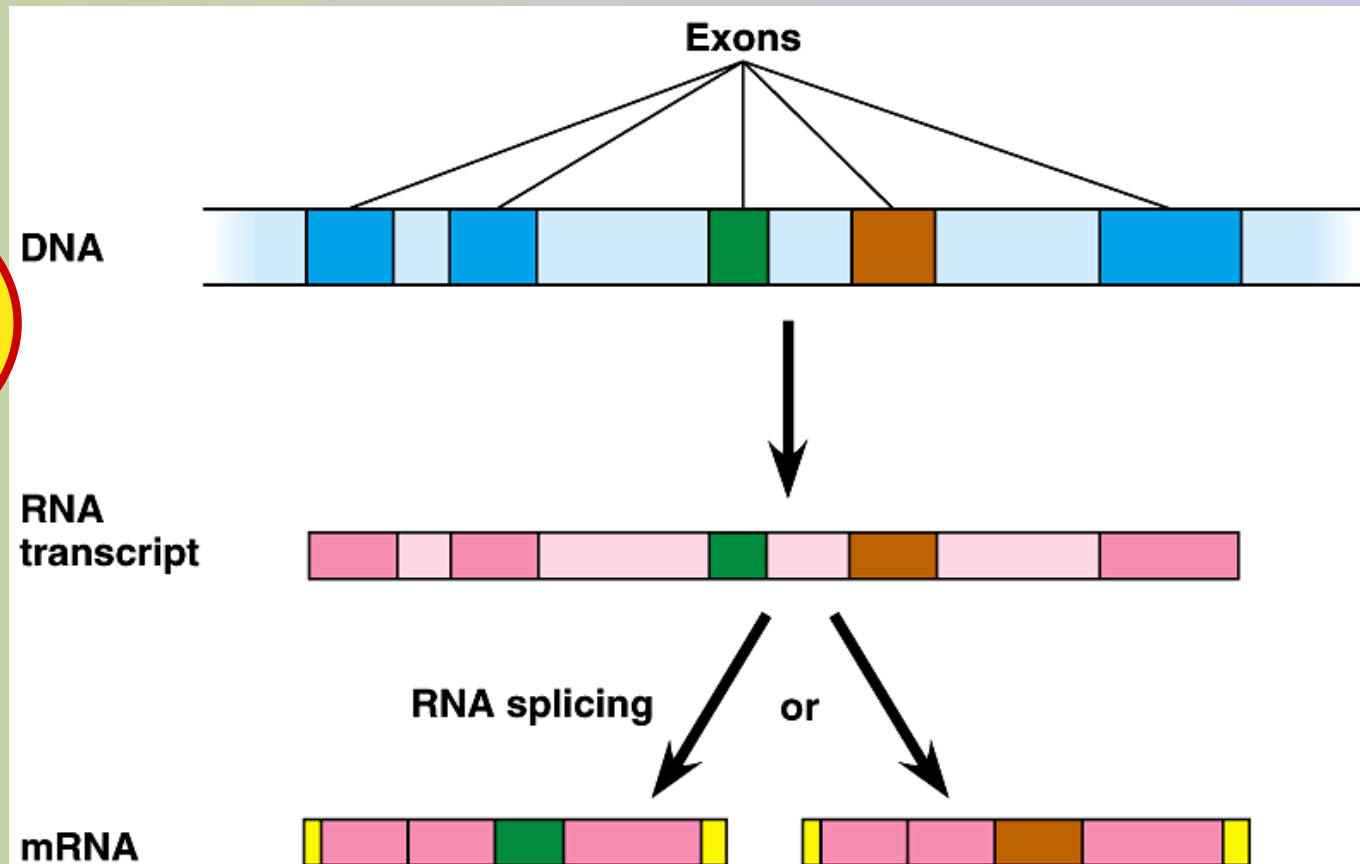
- snRNPs
  - small nuclear RNA
  - proteins
- Spliceosome
  - several snRNPs
  - recognize splice site sequence
    - cut & paste gene



# Alternative splicing

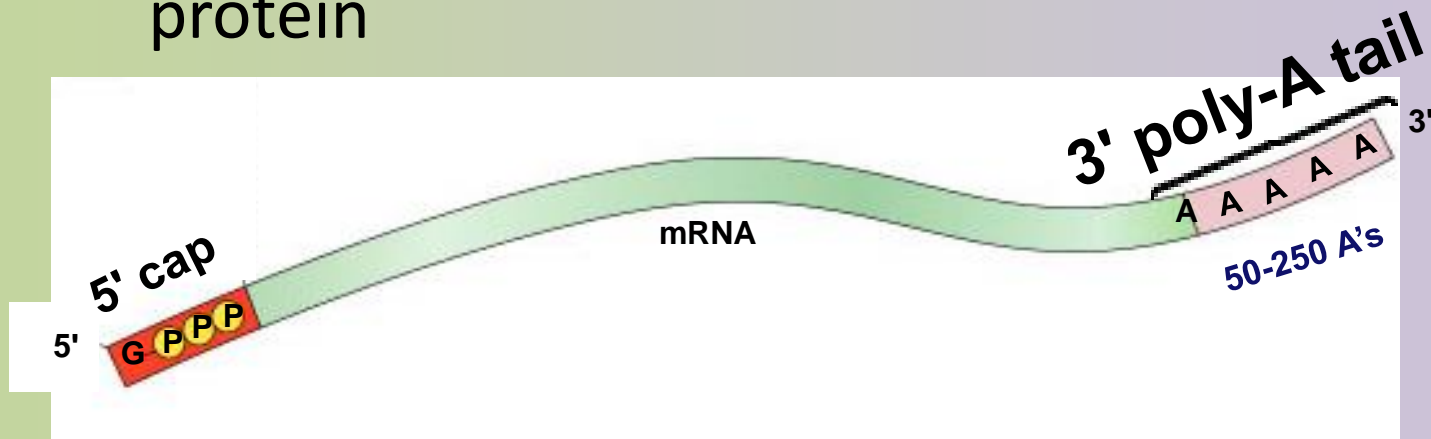
- Alternative mRNAs produced from same gene
  - when is an intron not an intron...
  - different segments treated as exons

Starting to get  
hard to  
define a gene!

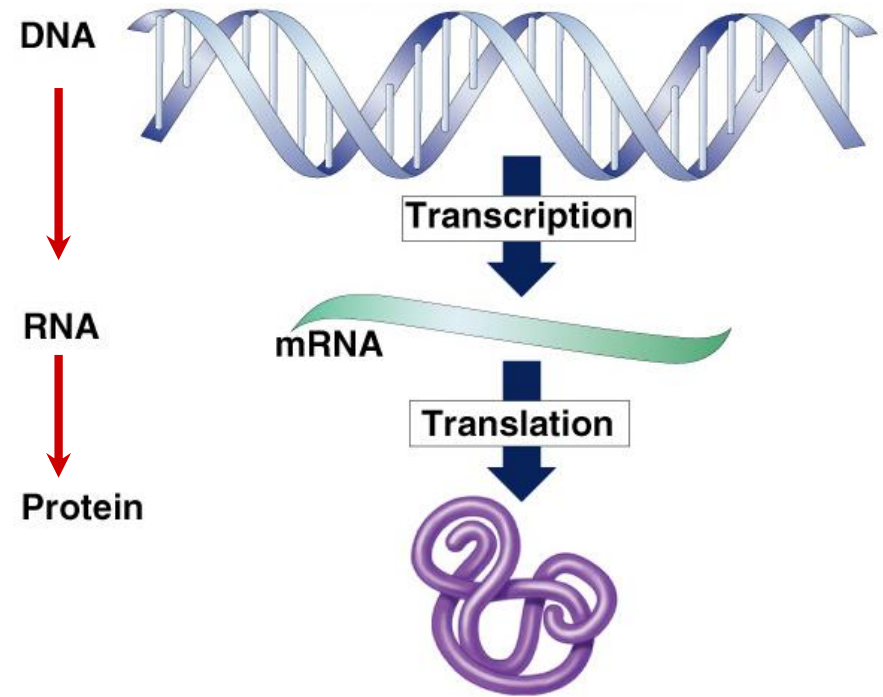


# More post-transcriptional processing

- Need to protect mRNA on its trip from nucleus to cytoplasm
  - enzymes in cytoplasm attack mRNA
    - protect the ends of the molecule
    - add 5' GTP cap
    - add poly-A tail
      - longer tail, mRNA lasts longer: produces more protein







## Translation 13.2

From nucleic acid language  
to amino acid language

# How does mRNA code for proteins?

**DNA**

4

ATCG

TACGCACATTTACGTACGCGG



**mRNA**

4

AUCG

AUGCGUGUAAAUGCAUGCGCC



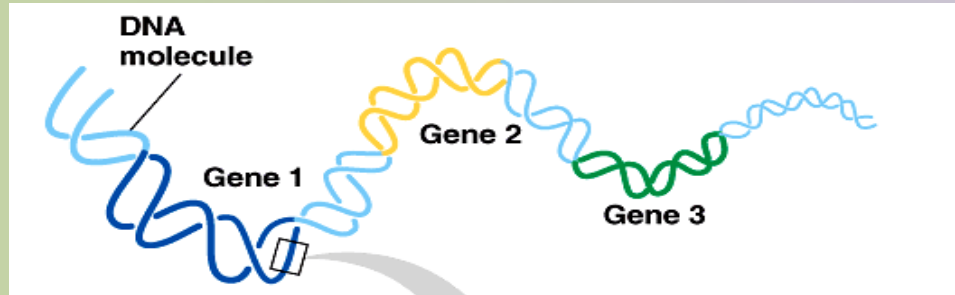
**protein**

20

Met Arg Val Asn Ala Cys Ala

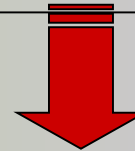
How can you code for 20 amino acids with only 4 nucleotide bases (A,U,G,C)?

# mRNA codes for proteins in triplets



**DNA**

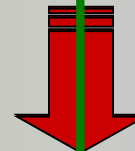
**TACGCACATTTACGTACGCGG**



**codon**

**mRNA**

**AUGCGUGUA~~AA~~UGCAUGCGCC**



**protein**

**Met Arg Val Asn Ala Cys Ala**

# The code

- Code for ALL life!
  - strongest support for a common origin for all life
- Code is redundant
  - several codons for each amino acid
  - 3rd base “wobble”

Why is the wobble good?

## ■ Start codon

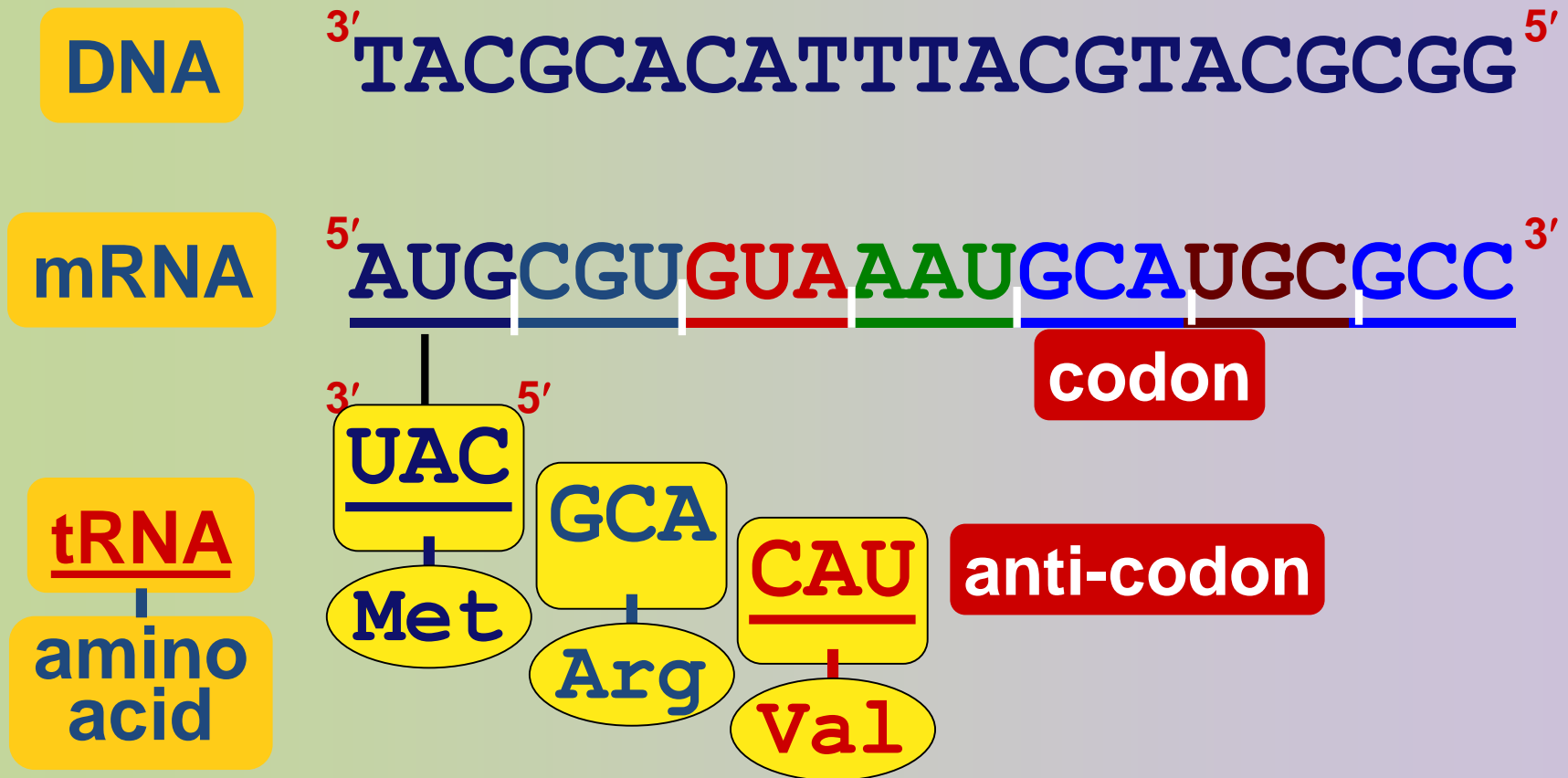
- ◆ AUG
- ◆ methionine

## ■ Stop codons

- ◆ UGA, UAA, UAG

		Second base				
		U	C	A	G	
First base (5' end)	U	UUU] Phe	UCU] Ser	UAU] Tyr	UGU] Cys	U
		UUC] Phe	UCC] Ser	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 or start	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
						Third base (3' end)

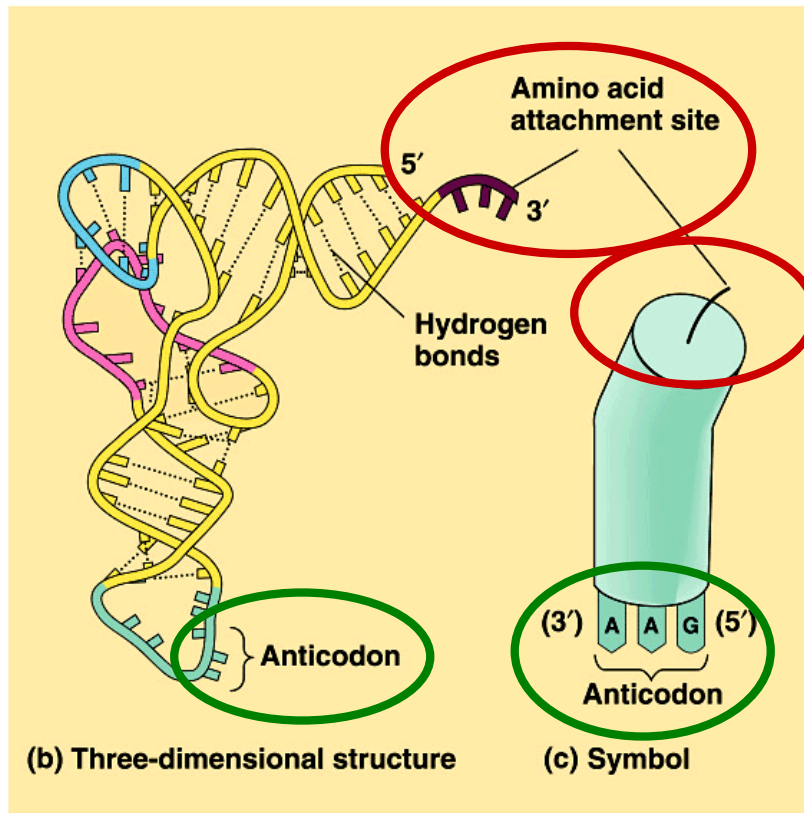
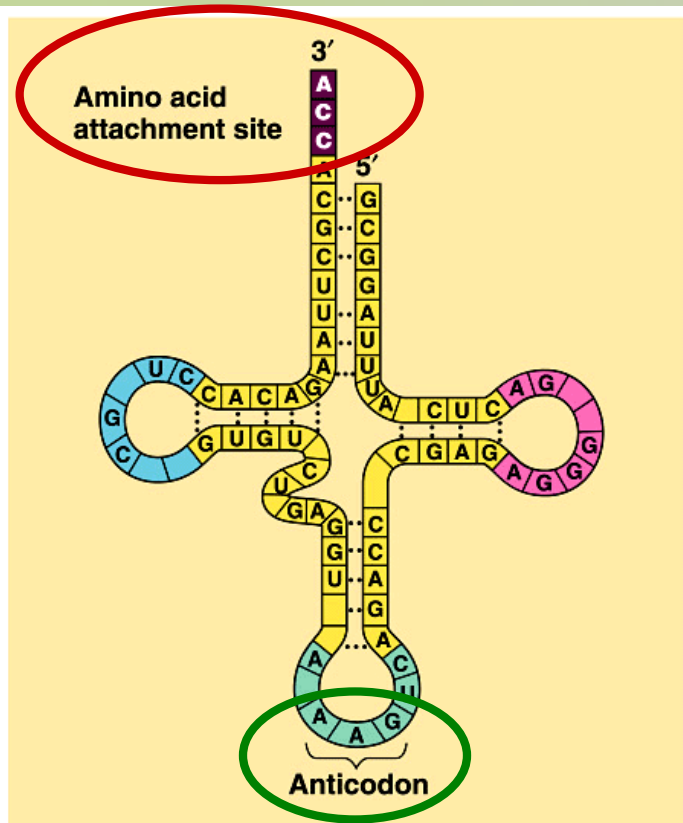
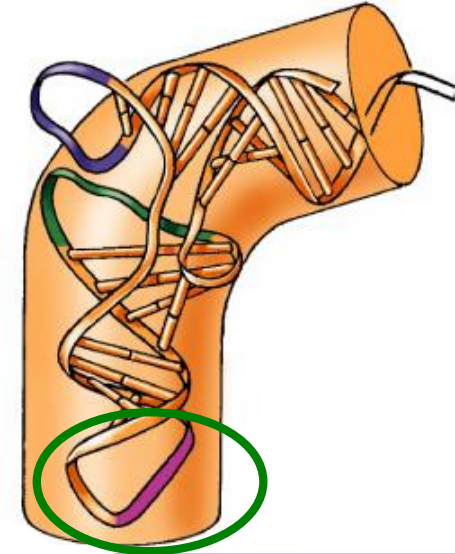
# How are the codons matched to amino acids?





# Transfer RNA structure

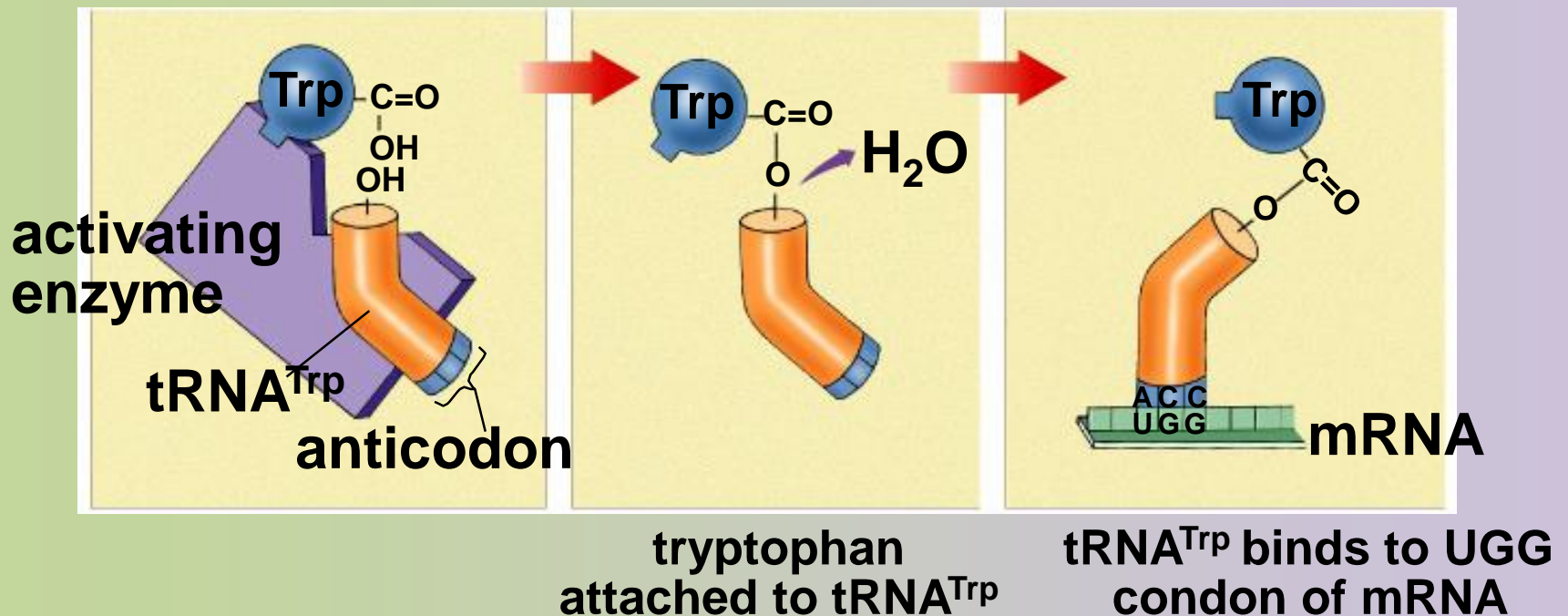
- “Clover leaf” structure
  - anticodon on “clover leaf” end
  - amino acid attached on 3' end



# Loading tRNA

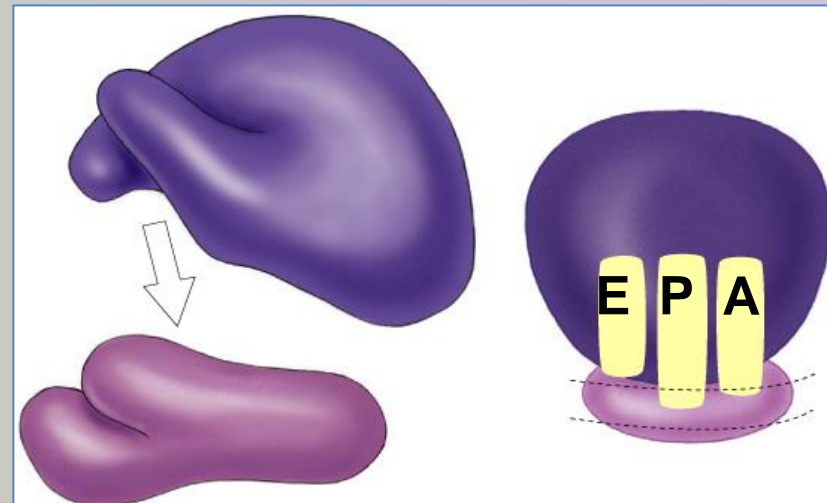
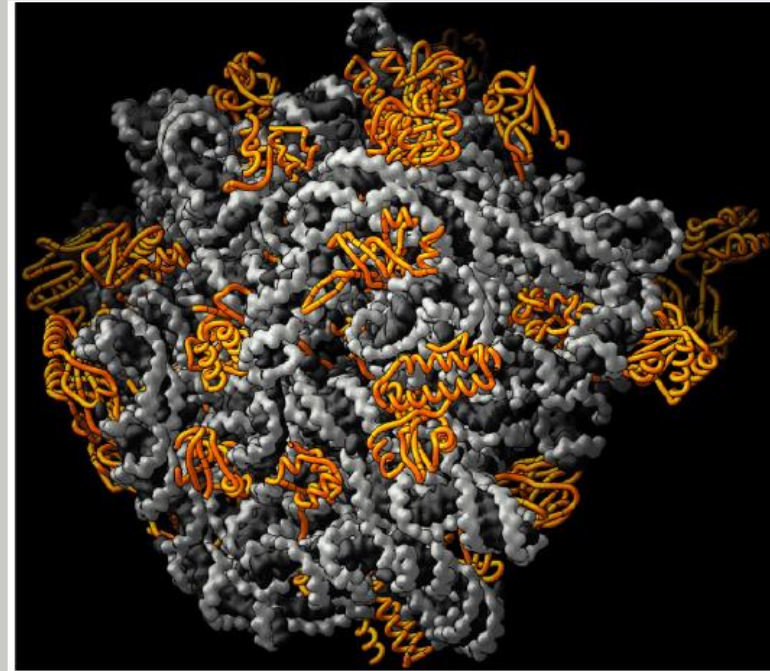
- Aminoacyl tRNA synthetase

- enzyme which bonds amino acid to tRNA
- bond requires energy
  - $\text{ATP} \rightarrow \text{AMP}$
  - bond is unstable
  - so it can release amino acid at ribosome easily



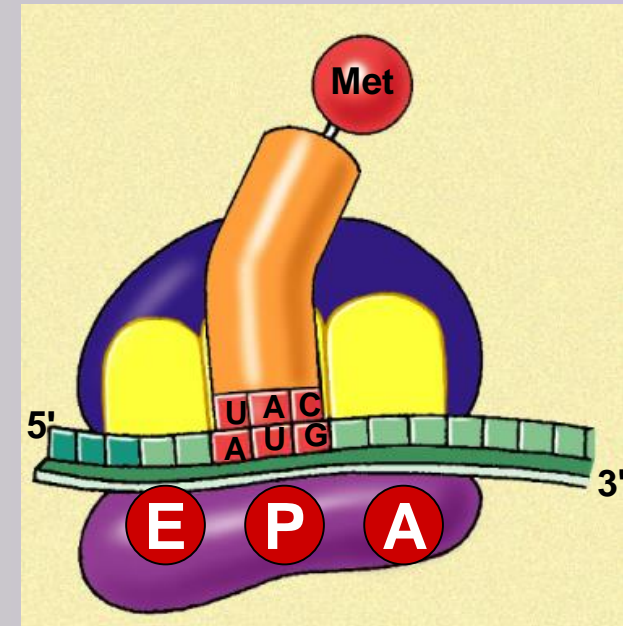
# Ribosomes

- Facilitate coupling of tRNA anticodon to mRNA codon
  - organelle or enzyme?
- Structure
  - ribosomal RNA (rRNA) & proteins
  - 2 subunits
    - large
    - small



# Ribosomes

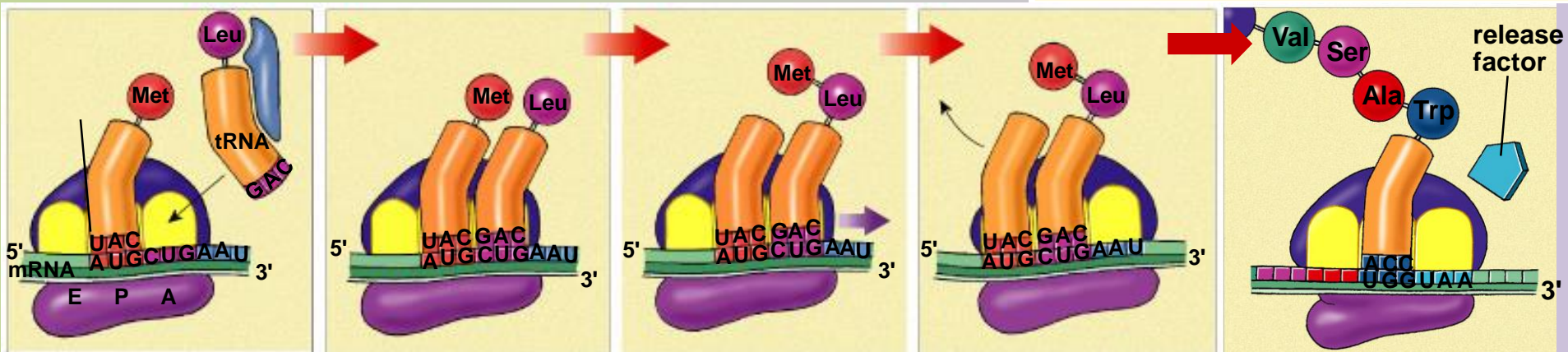
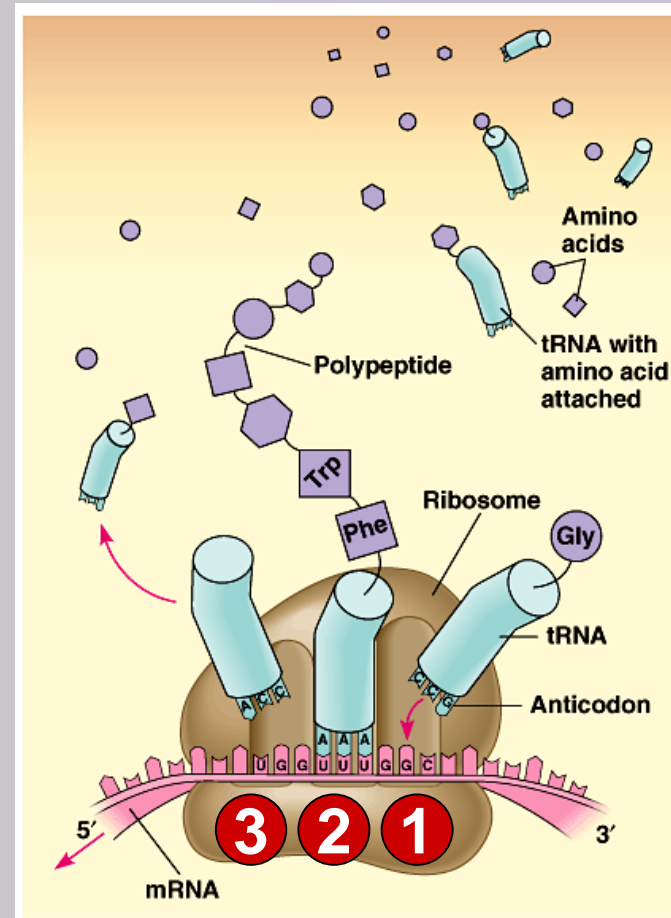
- A site (aminoacyl-tRNA site)
  - holds tRNA carrying next amino acid to be added to chain
- P site (peptidyl-tRNA site)
  - holds tRNA carrying growing polypeptide chain
- E site (exit site)
  - empty tRNA leaves ribosome from exit site





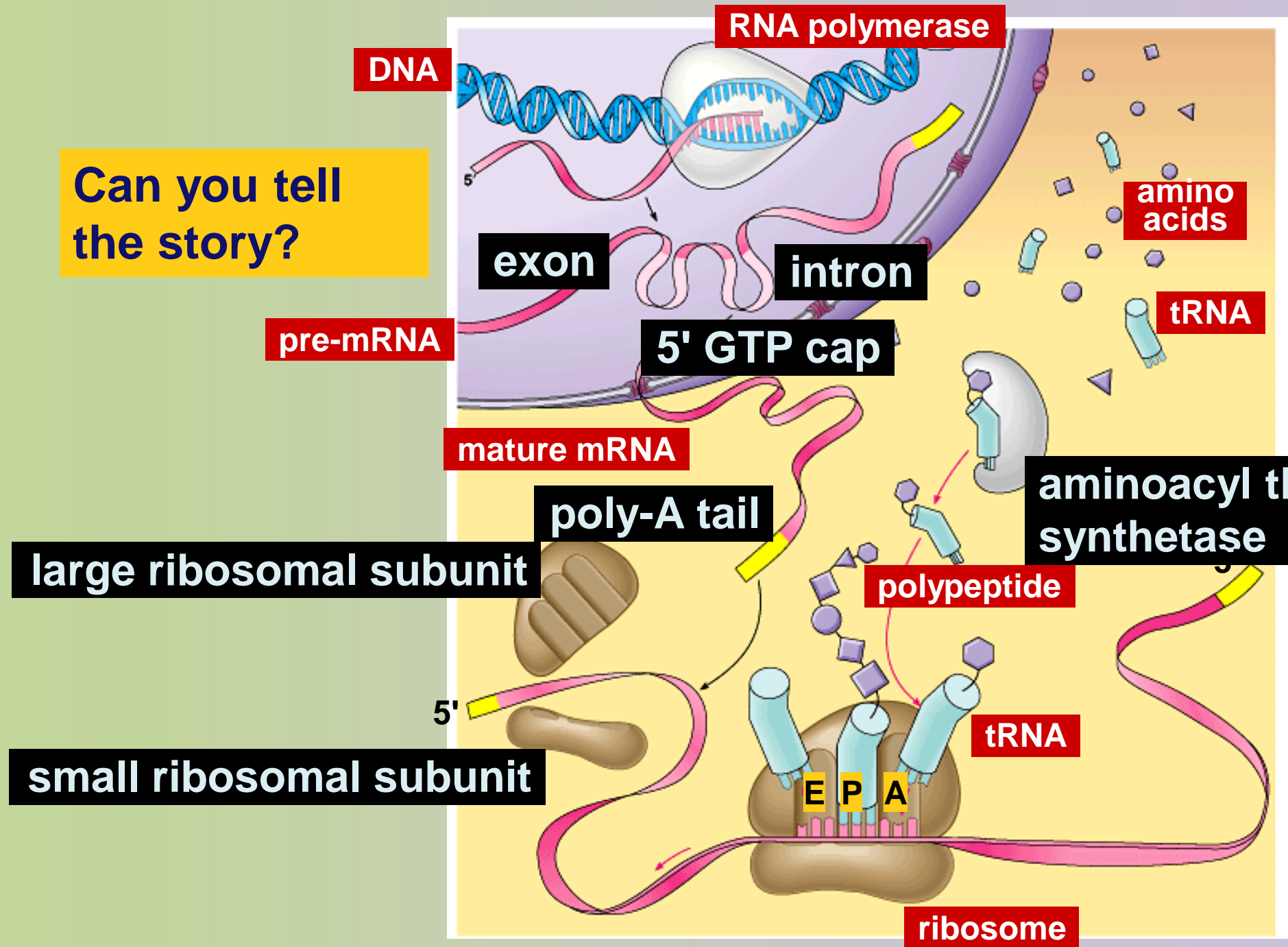
# Building a polypeptide

- Initiation
  - mRNA, ribosome subunits, initiator tRNA come together
- Elongation
  - adding amino acids based on codons
- Termination
  - STOP codon = Release factor

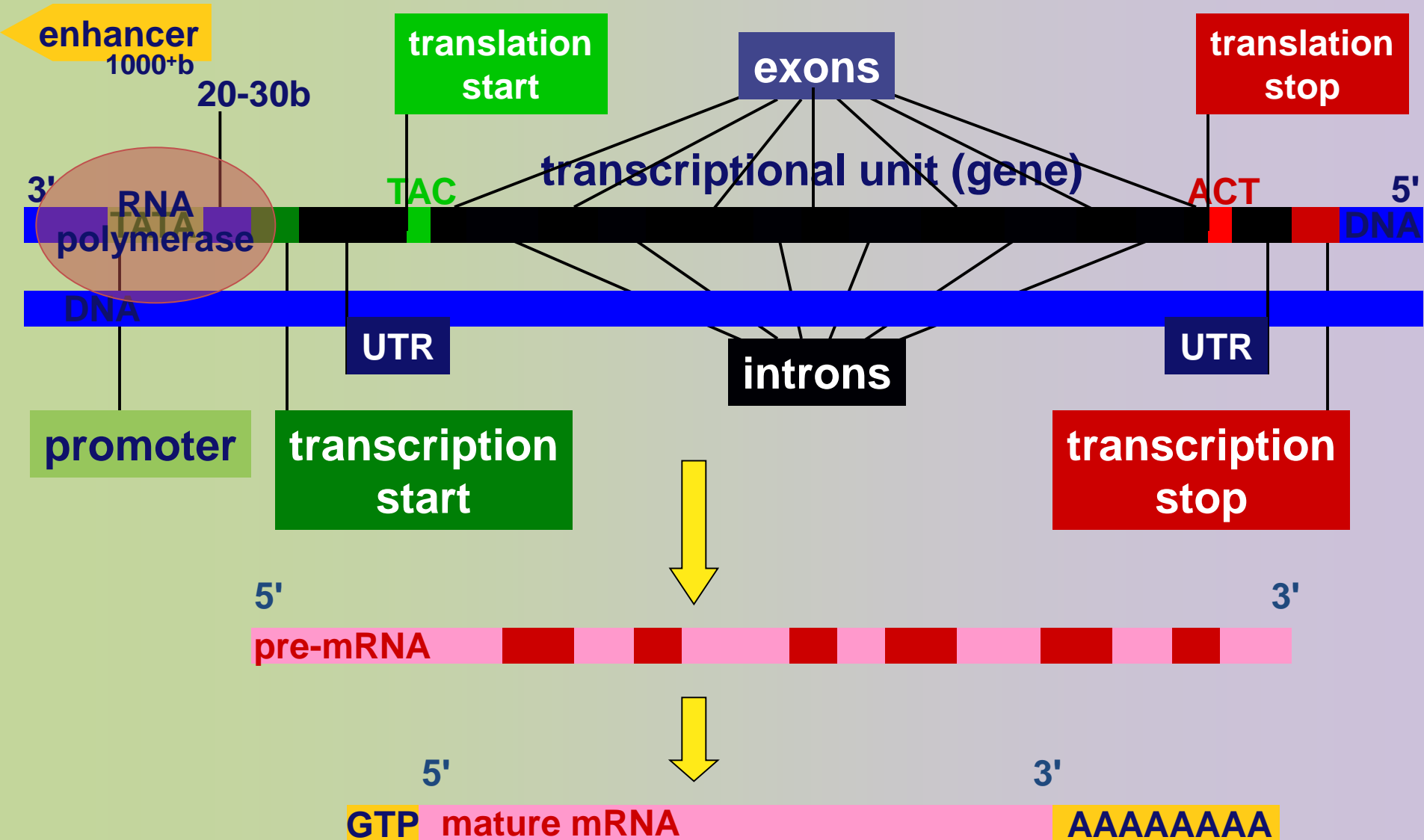




Can you tell the story?



# The Transcriptional unit (gene?)



# Protein Synthesis in Prokaryotes

Psssst...  
no nucleus!

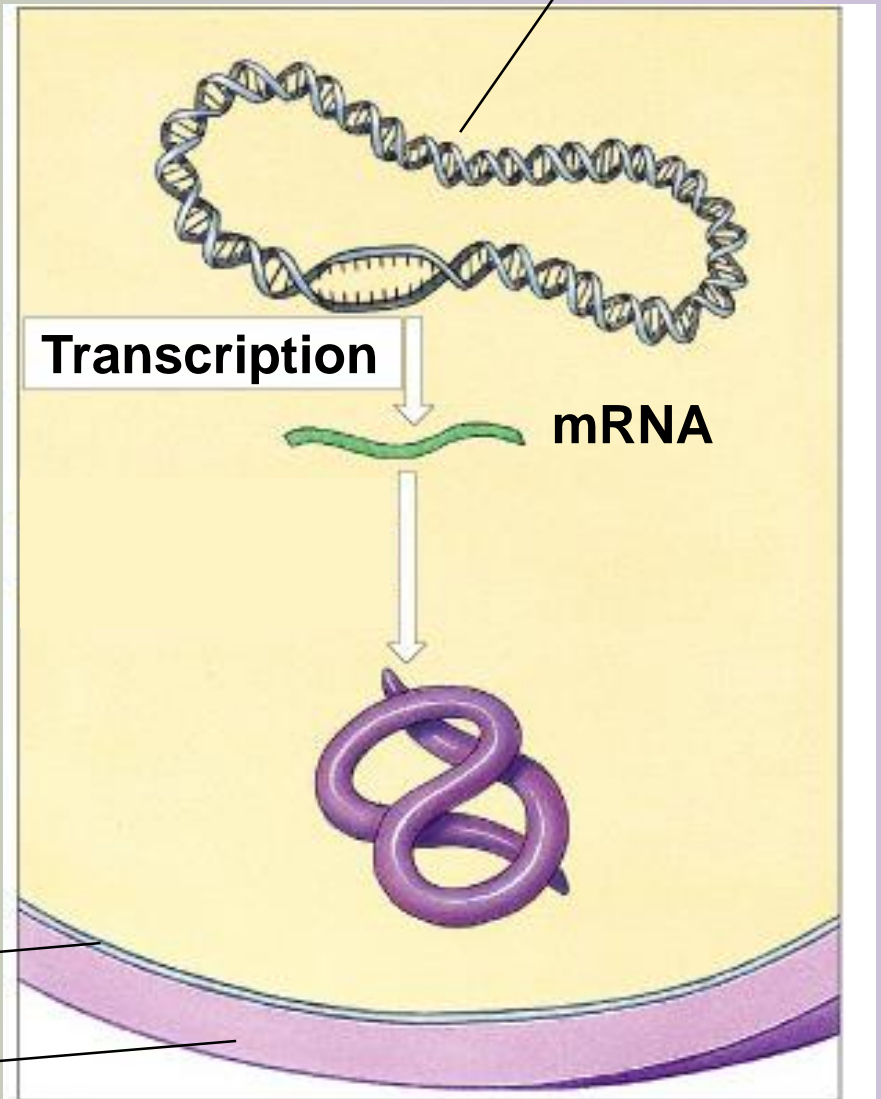
Cell  
membrane

Cell wall

Bacterial chromosome

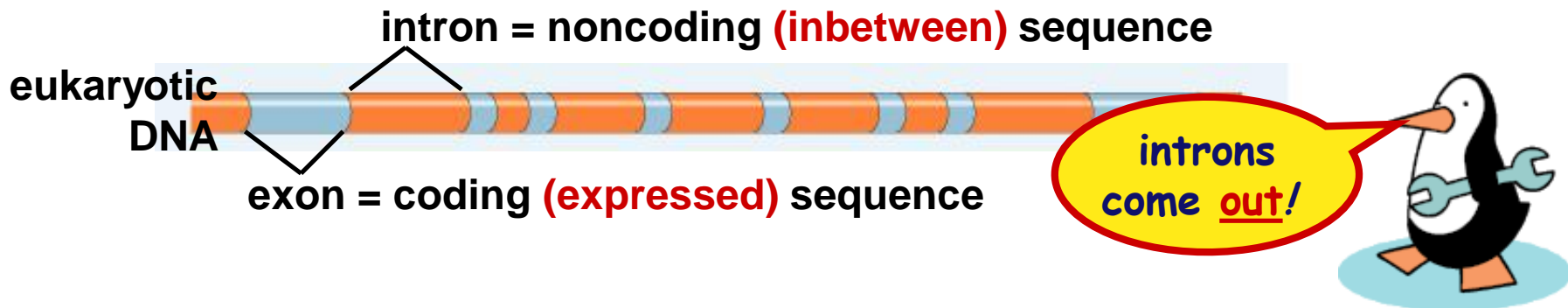
Transcription

mRNA



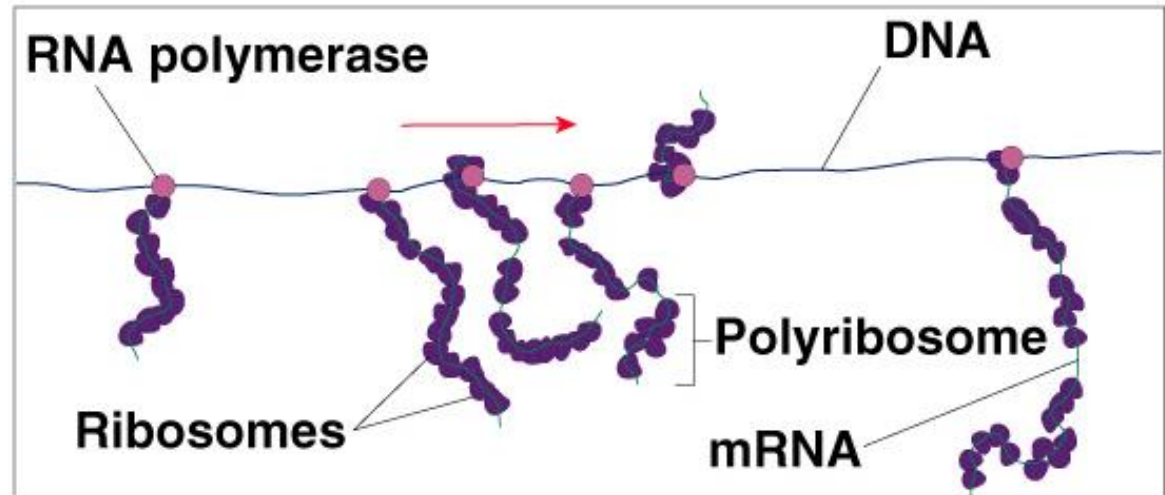
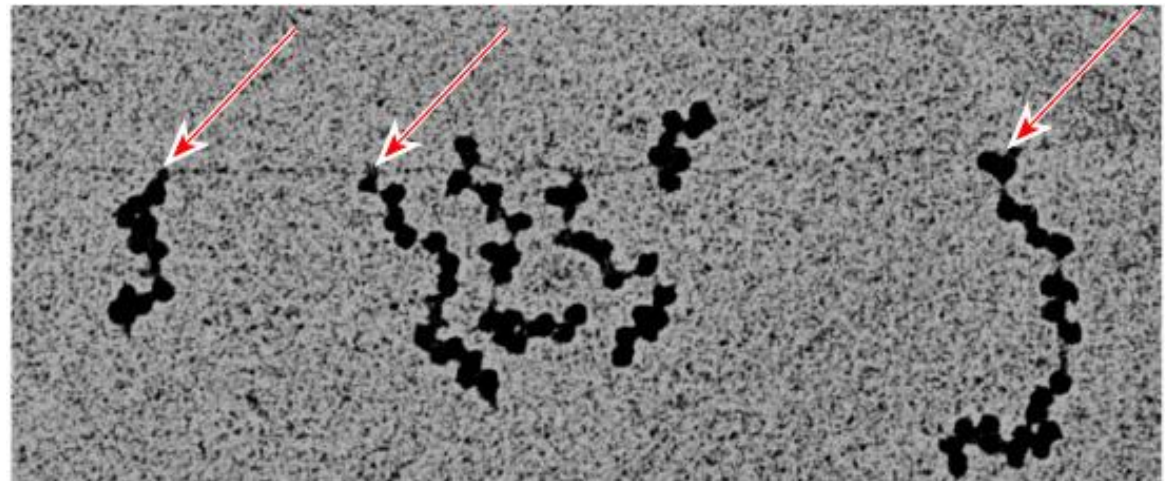
# Prokaryote vs. Eukaryote genes

- Prokaryotes
  - DNA in cytoplasm
  - circular chromosome
  - naked DNA
  - no introns
- Eukaryotes
  - DNA in nucleus
  - linear chromosomes
  - DNA wound on histone proteins
  - introns vs. exons



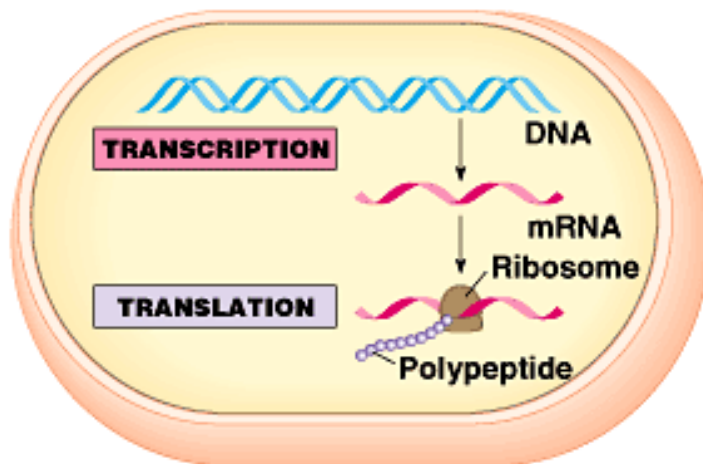
# Translation in Prokaryotes

- Transcription & translation are simultaneous in bacteria
  - DNA is in cytoplasm
  - no mRNA editing
  - ribosomes read mRNA as it is being transcribed

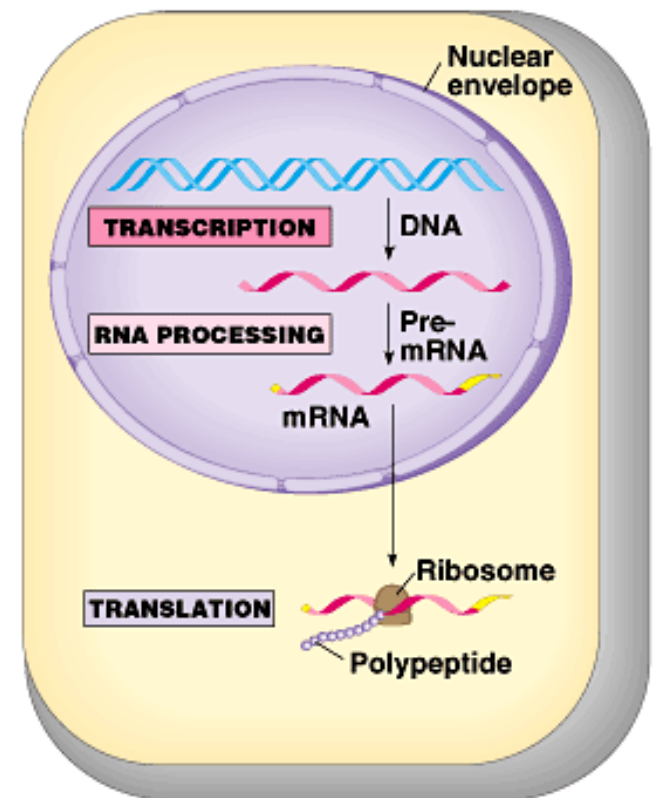


# Translation: prokaryotes vs. eukaryotes

- Differences between prokaryotes & eukaryotes
  - time & physical separation between processes
    - takes eukaryote ~1 hour from DNA to protein
  - no RNA processing

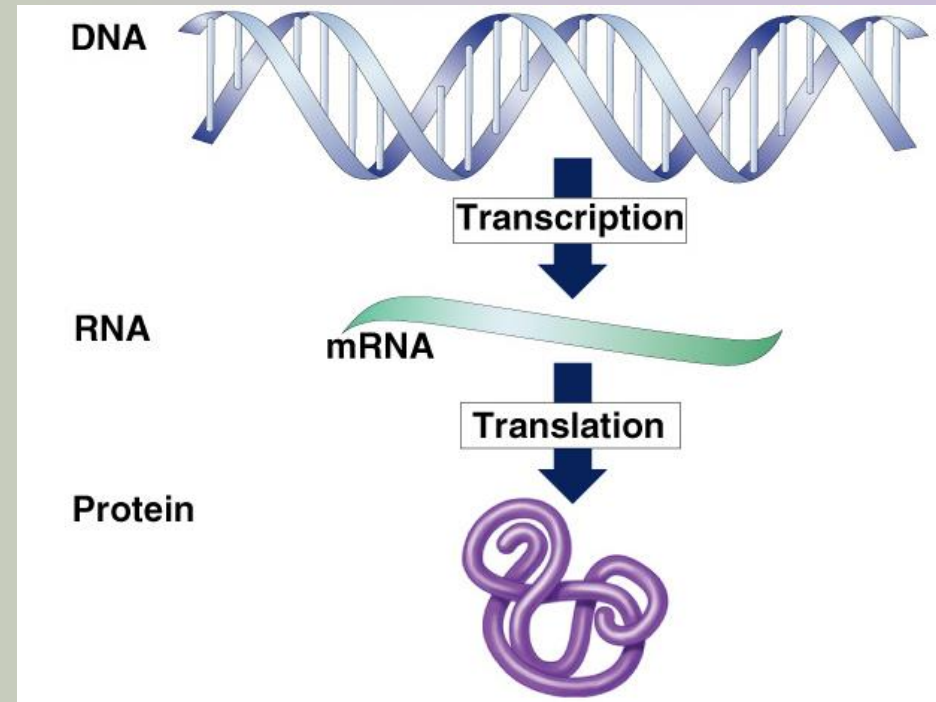


(a) Prokaryotic cell



(b) Eukaryotic cell





## Mutations (Ch. 13.3)



10  $\mu$ m

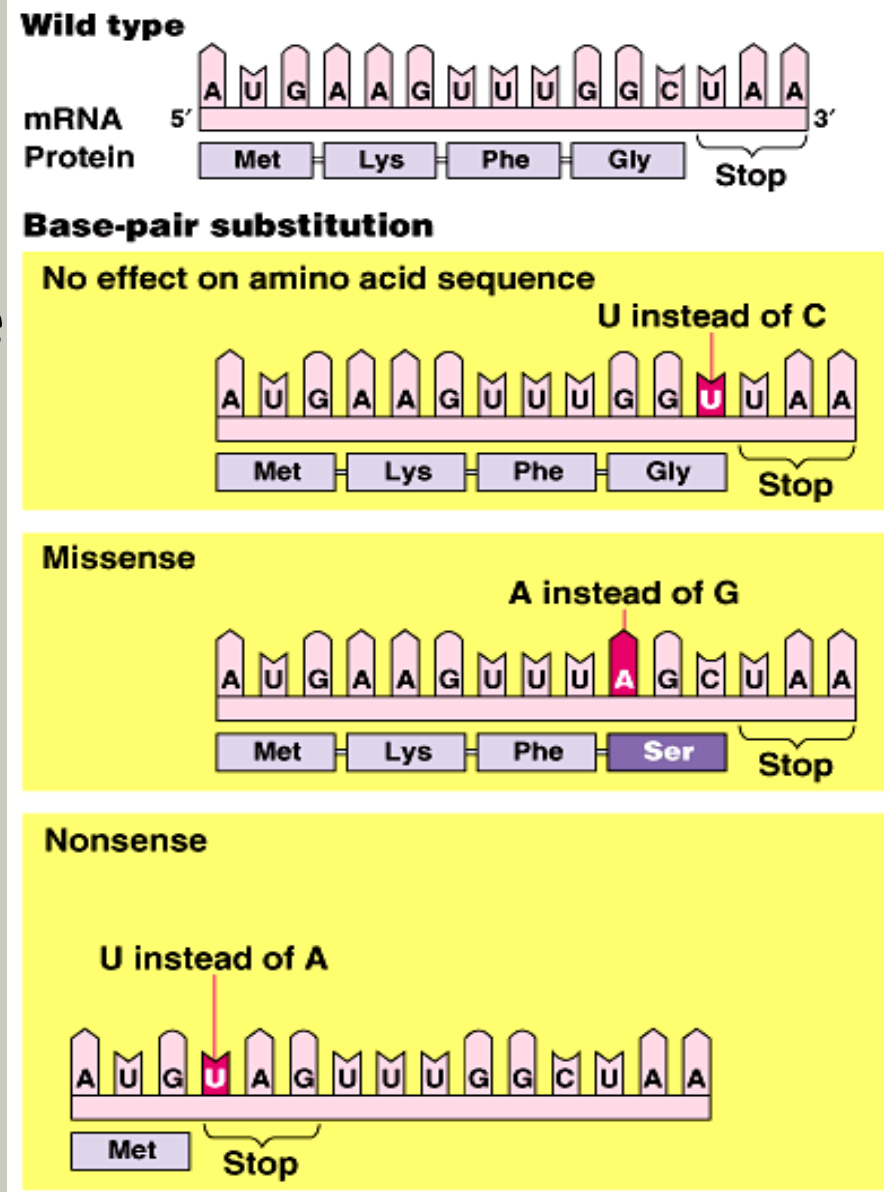
Val	His	Leu	Thr	Pro	Val	Glu	...
1	2	3	4	5	6	7	

(b) Sickled red blood cells and the primary structure of sickle-cell hemoglobin



# Mutations

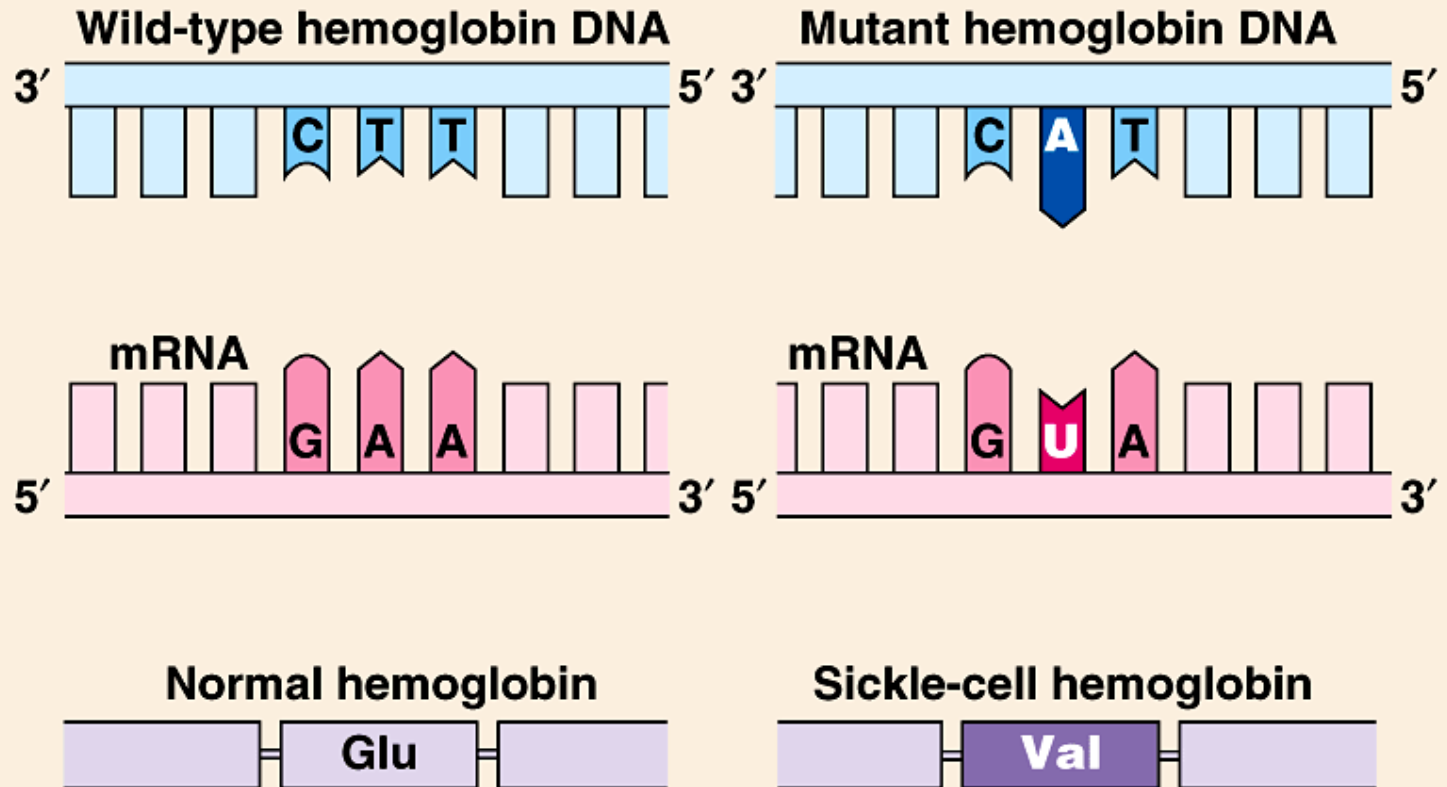
- Point mutations
  - single base change
    - silent mutation
      - no amino acid change
      - redundancy in code
    - missense
      - change amino acid
    - nonsense
      - change to stop codon



When do mutations  
affect the next  
generation?

# Point mutation lead to Sick cell anemia

What kind of mutation?



**Missense!**



# Mutations

- Frameshift

- shift in the reading frame

- changes everything “downstream”

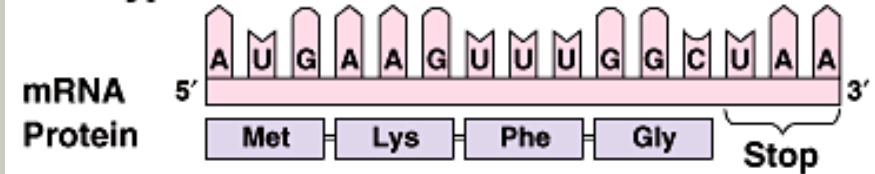
- insertions

- adding base(s)

- deletions

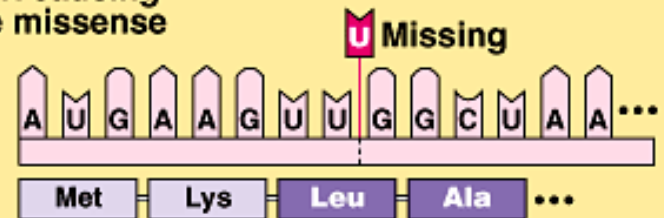
- losing base(s)

## Wild type

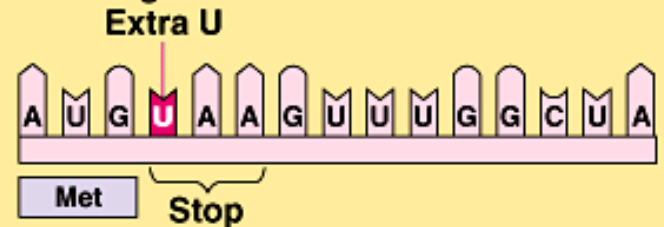


## Base-pair insertion or deletion

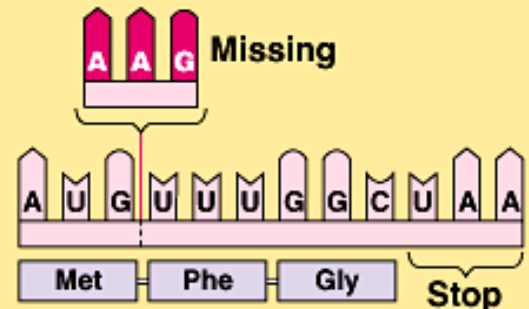
### Frameshift causing extensive missense



### Frameshift causing immediate nonsense

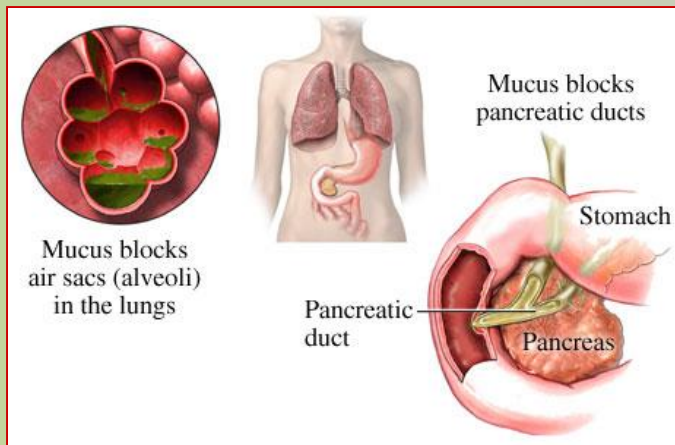


### Insertion or deletion of 3 nucleotides: no frameshift; extra or missing amino acid



Where would this mutation cause the most change: beginning or end of gene?

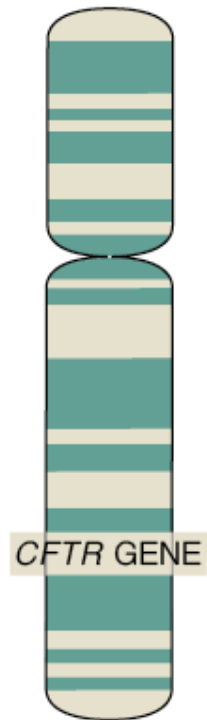




Chromosome 7

Sequence of nucleotides in *CFTR* gene

Amino acid sequence of CFTR protein



A  
T  
C

— ISOLEUCINE 506

A  
T

— ISOLEUCINE 507

**C**  
**T**  
**T**

— **PHENYLALANINE 508**

T

G  
G  
T

— GLYCINE 509

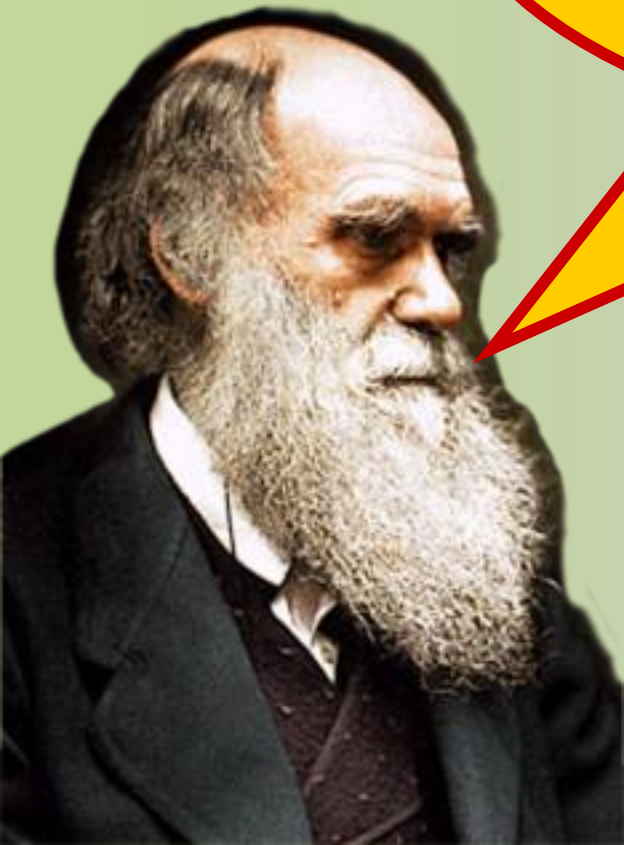
G  
T  
T

— VALINE 510

**DELETED IN MANY PATIENTS WITH CYSTIC FIBROSIS**



**What's the value of  
mutations?**



# Review Questions

1. If proteins were composed of only 12 different kinds of amino acids, what would be the smallest possible codon size in a genetic system with four different nucleotides?

- A. 1
- B. 2
- C. 3
- D. 4
- E. 12



2. A portion of the genetic code is UUU = phenylalanine, GCC = alanine, AAA = lysine, and CCC = proline. Assume the correct code places the amino acids phenylalanine, alanine, and lysine in a protein (in that order). Which of the following DNA sequences would substitute proline for alanine?

A. AAA-CGG-TTA

B. AAT-CGG-TTT

C. AAA-CCG-TTT

D. AAA-GGG-TTT

E. AAA-CCC-TTT

3. What is the relationship among DNA, a gene, and a chromosome?

- A. A chromosome contains hundreds of genes, which are composed of protein.
- B. A chromosome contains hundreds of genes, which are composed of DNA.
- C. A gene contains hundreds of chromosomes, which are composed of protein.
- D. A gene is composed of DNA, but there is no relationship to a chromosome.
- E. A gene contains hundreds of chromosomes, which are composed of DNA.

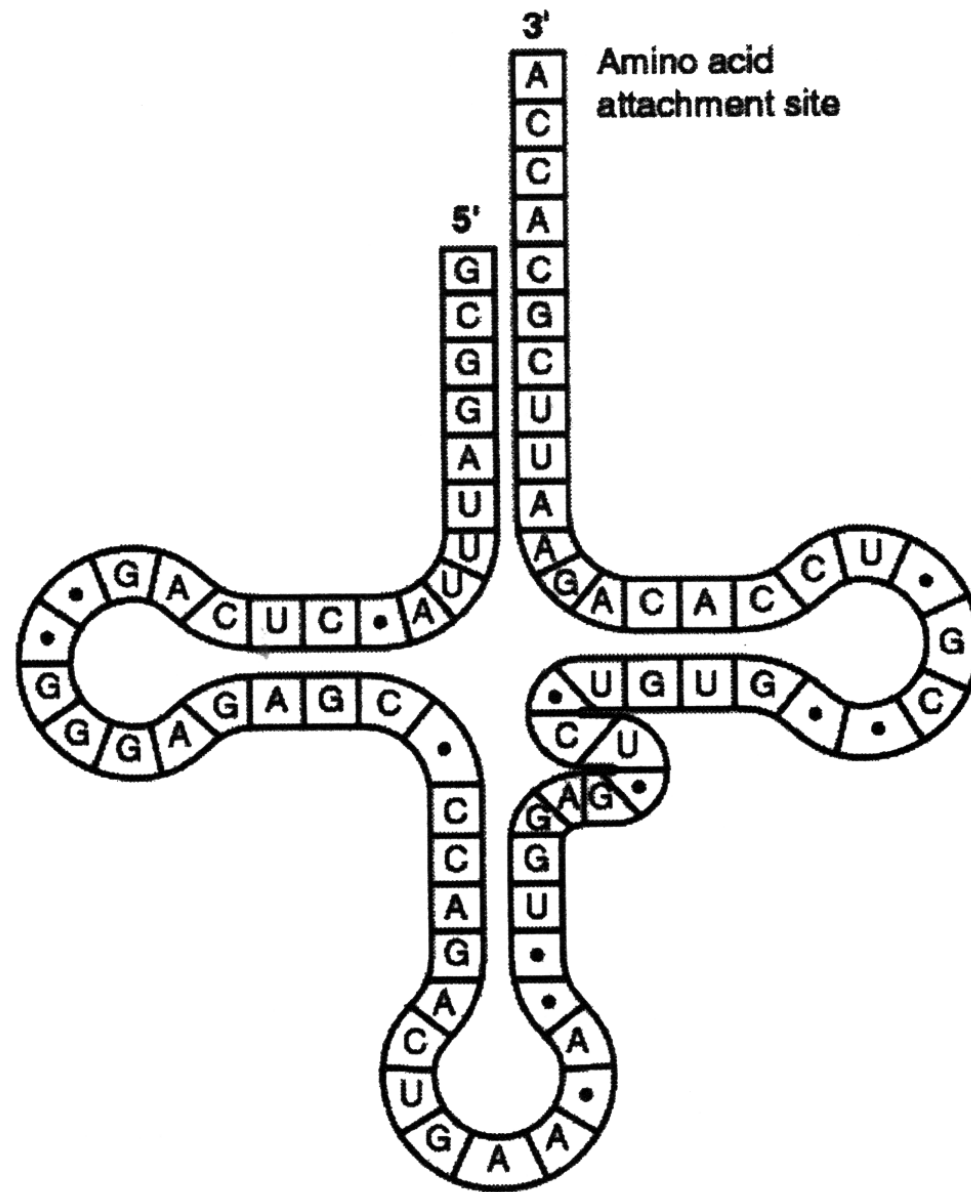
4. A particular triplet of bases in the coding sequence of DNA is AAA. The anticodon on the tRNA that binds the mRNA codon is
- A. TTT.
  - B. UUA.
  - C. UUU.
  - D. AAA.
  - E. either UAA or TAA, depending on wobble in the first base.

5. A part of an mRNA molecule with the following sequence is being read by a ribosome: 5' CCG-ACG 3' (mRNA). The following activated transfer RNA molecules are available. Two of them can correctly match the mRNA so that a dipeptide can form.

<b>tRNA Anticodon</b>	<b>Amino Acid</b>
GGC	Proline
CGU	Alanine
UGC	Threonine
CCG	Glycine
ACG	Cysteine
CGG	Alanine

The dipeptide that will form will be

- A. cysteine-alanine.
- B. proline-threonine.
- C. glycine-cysteine.
- D. alanine-alanine.
- E. threonine-glycine.



6. This figure represents tRNA that recognizes and binds a particular amino acid (in this instance, phenylalanine). Which of the following triplets of bases on the mRNA strand codes for this amino acid?

- A. UGG
- B. GUG
- C. GUA
- D. UUC
- E. CAU

7. How is the template strand for a particular gene determined?

- A. It is the DNA strand that runs from the 5' → 3' direction.
- B. It is the DNA strand that runs from the 3' → 5' direction.
- C. It depends on the orientation of RNA polymerase, whose position is determined by particular sequences of nucleotides within the promoter.
- D. It doesn't matter which strand is the template because they are complementary and will produce the same mRNA.
- E. The template strand always contains the TATA box.

8. A biologist inserts a gene from a human liver cell into the chromosome of a bacterium. The bacterium then transcribes this gene into mRNA and translates the mRNA into protein. The protein produced is useless. The biologist extracts the protein and mature mRNA that codes for it. When analyzed you would expect which of the following results? \*

- A. the protein and the mature mRNA are longer than in human cells
- B. the protein and mature mRNA are shorter than expected
- C. the protein is longer and the mRNA is shorter than expected
- D. the protein is shorter and the mRNA is longer than expected



9. If the structure of a TV show is analogous to the structure of a gene, then the introns of a gene would be analogous to
- A. the opening theme music.
  - B. the segments of the show.
  - C. the commercials between segments of the show.
  - D. the commercials between shows.
  - E. the closing credits.

10. Each of the following is a modification of the sentence THECATATETHERAT.

- A. *THERATATETHECAT*
- B. *THETACATETHERAT*
- C. *THECATARETHERAT*
- D. *THECATATTHERAT*
- E. *CATATETHERAT*

Which of the above is analogous to a frameshift mutation?

- A
- B
- C
- D
- E

11. Each of the following is a modification of the sentence THECATATETHERAT.

- A. *THERATATETHECAT*
- B. *THETACATETHERAT*
- C. *THECATARETHERAT*
- D. *THECATATTHERAT*
- E. *CATATETHERAT*

Which of the above is analogous to a single substitution mutation?

- A
- B
- C
- D
- E