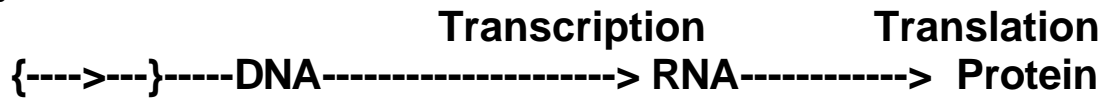


Lecture Summary:

We have been talking about the flow of information in Biological Systems.



Replication

Now we will begin looking at the last step in this flow of information.

Translation of Messenger RNA into Protein

As we discussed above, the flow of Information is from DNA to RNA to Protein.



Information is stored in the form of linear nucleotide sequence in DNA.

DNA is transcribed into Messenger RNA:



But Information is still in the form of a linear nucleotide sequence.

Messenger RNA is then translated to make a protein with a specific linear amino acid sequence:

Met-Asp-Leu-Arg-Val-Trp-Asp-Thr-Lys-Gly-Tyr- etc.

We can see that the information has changed form.

Information is transformed from nucleotide to amino acid sequence.

How is this accomplished?

You can imagine this process is similar to translating a book from one language to another.

This is why this process is called "Translation".
As we have seen, amino acids are the words in the "protein language".

But how are amino acid sequences written in DNA and mRNA nucleotide sequences?

Language of nucleotide sequences is called **The Genetic Code**.

Genetic Code is written in words containing three nucleotides each.

These three nucleotide words are called "**Codons**".

Each codon has meaning. A **CODON** is used to indicate a single amino acid or to indicate the termination of protein synthesis.

The genetic code is therefore the relationship between the sequence of bases in a DNA gene (or its RNA transcript) and the sequence of amino acids in the protein that the gene encodes.

Features of the Genetic Code.

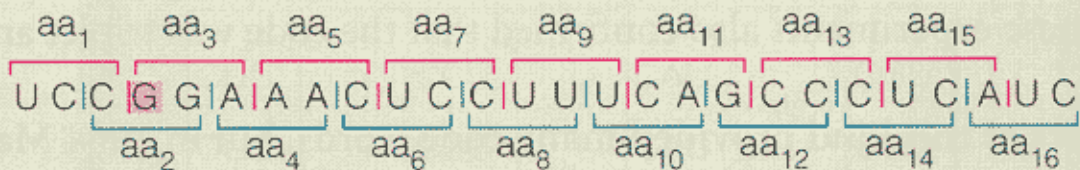
1) **The coding Ratio:** There are twenty different common amino acids, but only four different bases in a mRNA molecule.

A single base code can only specify 4 kinds of amino acids because DNA only has four kinds of bases.

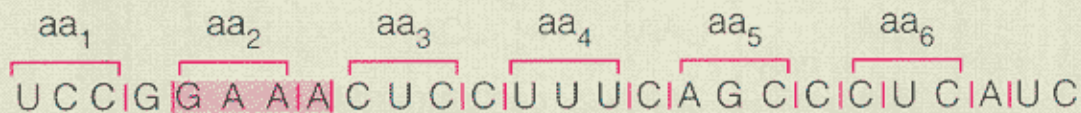
A two-base code would allow 16 ($4 \times 4 = 16$) while a three base code would allow 64 ($4 \times 4 \times 4 = 64$).

Since 20 common amino acids are found in proteins, the genetic code must be base 3 or greater (in fact: An amino acid is encoded for by a group of three bases / these three bases constitute a codon).

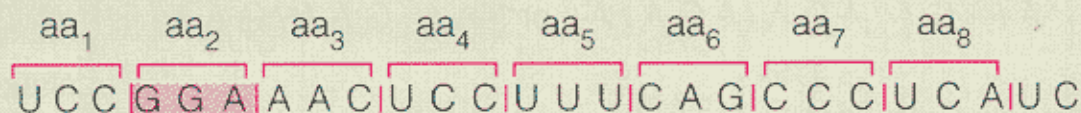
2) **Code overlapping or nonoverlapping?**



(a) Overlapping code. There will be statistical regularities between adjacent amino acid residues. Point mutations (red) will be able to change two amino acid residues.



(b) Punctuated code. Deletions of four nucleotides (or multiples thereof) will restore the reading frame.



(c) Unpunctuated code. Deletions of three nucleotides (or multiples thereof) will restore the reading frame. This is the actual form of the code.

3) How is the correct group of bases read? Sequence of bases is read sequentially from a fixed starting point.

^ start point
 ABC DEF GHI JKL
 aa1 aa2 aa3 aa4

If a mutation results in the loss of a single base (Lose G):

Mutant strand: ABC DEF HIJ KL.....
 aa1 aa2 aa3* aa4*

Termed a Deletion Mutant.

If a mutation results in the gain of a single base (gain Z):

Mutant Strand: ABC DZE FGH IJK LM.....
 aa1 aa2 aa3 aa4 aa5

Termed an Insertion Mutant.

4) There are 64 codons but only 20 amino acids. 61 of the 64 codons code for amino acids (the other codons encode stop codons). Thus, most amino acids are coded for by more than one codon (the code is degenerate).

First position (5' end)	Second position				Third position (3' end)
	U	C	A	G	
U	Phe	Ser	Tyr	Cys	U
	Phe	Ser	Tyr	Cys	C
	Leu	Ser	STOP	STOP	A
	Leu	Ser	STOP	Trp	G
C	Leu	Pro	His	Arg	U
	Leu	Pro	His	Arg	C
	Leu	Pro	Gln	Arg	A
	Leu	Pro	Gln	Arg	G
A	Ile	Thr	Asn	Ser	U
	Ile	Thr	Asn	Ser	C
	Ile	Thr	Lys	Arg	A
	Met	Thr	Lys	Arg	G
G	Val	Ala	Asp	Gly	U
	Val	Ala	Asp	Gly	C
	Val	Ala	Glu	Gly	A
	Val	Ala	Glu	Gly	G

Points about Genetic Code

1. Code is said to be "**Degenerate**". Most amino acids have more than one codon.

Leu can be designated by CUU, CUC, CUA, or CUG (short hand CUN).

Histidine coded by both CAU and CAC (short hand: CAU/C).

This is one reason why not all mutations in DNA cause problems. In many cases, mutation in the third position of codons causes no change in the amino acid that is coded.

2. **Each codon stands for only one amino acid.** Has only one meaning.

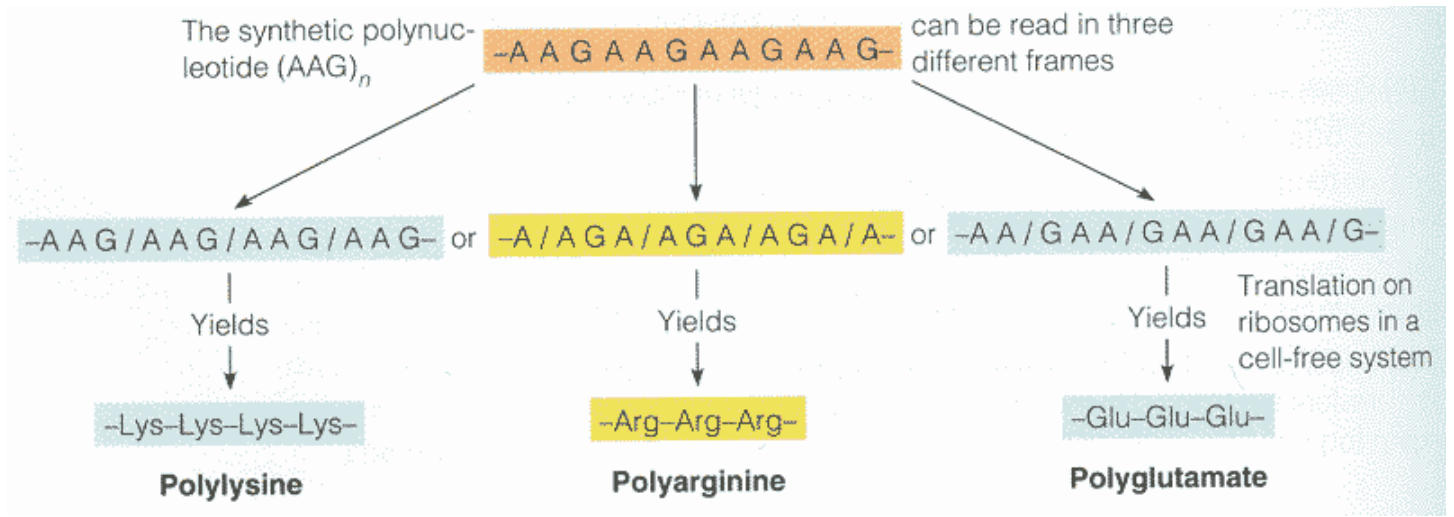
3. The codons UAA, UAG, and UGA don' t code for amino acids. They are "**Termination Codons**".

Tell the ribosome to stop translating the mRNA and terminate translation.

The Genetic Code allows you to determine what amino acid sequence is coded for by a particular nucleotide sequence in mRNA.

Example:

5'AUG/GAT/CTG/GCC/GTG.... 3'.
Met Asp Leu Ala Val



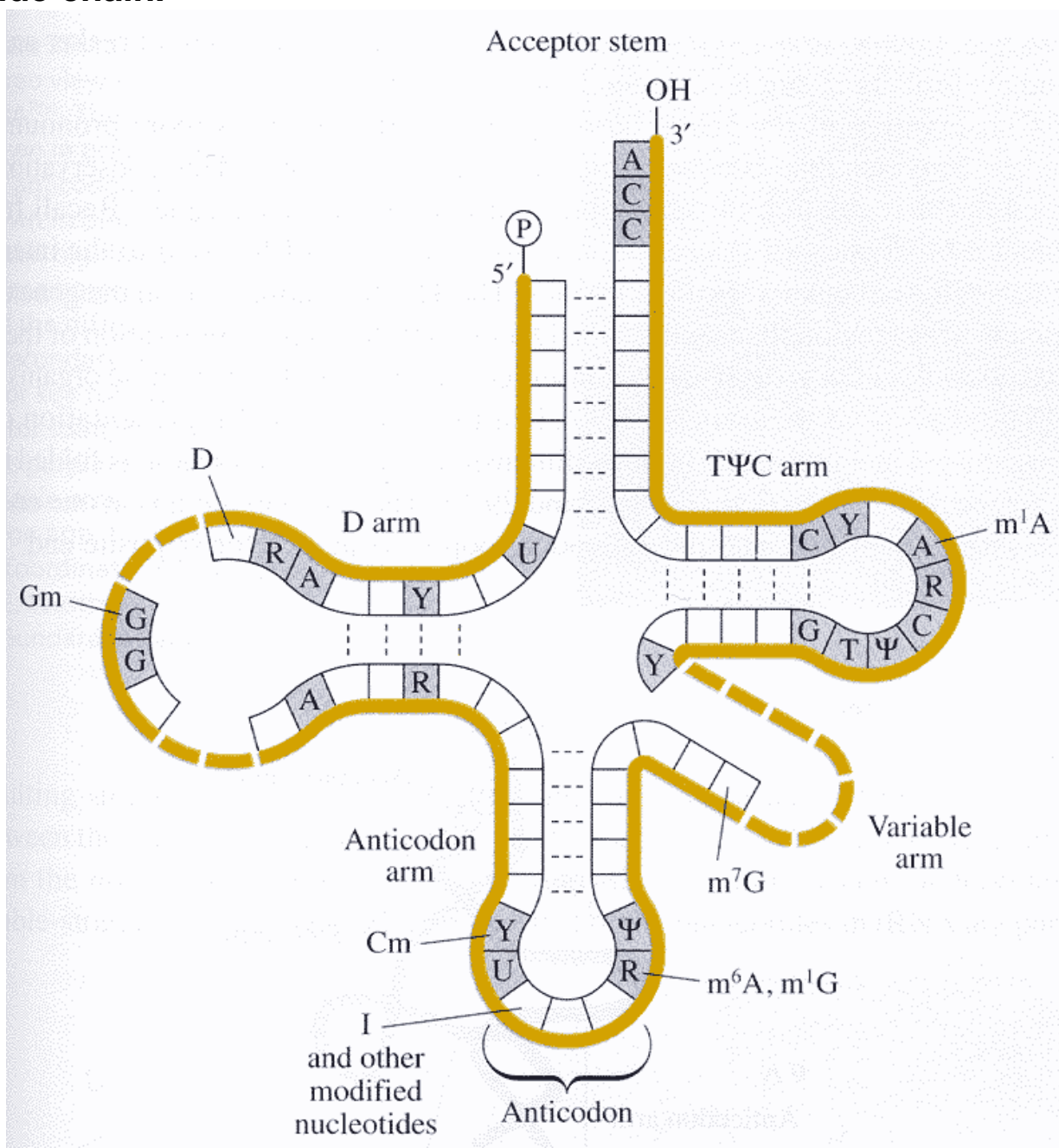
But when ribosomes translate mRNA how are nucleotide codons converted to an amino acid sequence?

This is the function of **Transfer RNAs (tRNAs)**.

There are more than 20 different tRNAs.

The function of tRNA's:

- 1) To chemically link to a particular amino acid (covalent linkage).
- 2) To recognize a specific codon in mRNA (non-covalent) so that the attached amino acid can be added to the growing peptide chain.



General Structure of Transfer RNAs:

- 1) Single polynucleotide chains containing 73-93 ribonucleotides
- 2) They contain many unusually modified bases. (Inosine).
- 3) The 5' end of tRNA is phosphorylated, and the 5' residue is usually pG.
- 4). The base sequence at the 3' end of a mature tRNA is **CCA**. The activated amino acid is attached to the 3'-OH group of the adenosine.
- 5) About half of the nucleotides in tRNAs are base-paired to form double stranded helices.
- 6) The anticodon loop consists of seven bases, with this sequence:

5' -pyrimidine-pyrimidine-X-Y-Z-modified purine- variable base- 3'

We will focus on two portions of the tRNA: Anticodon and Amino Acid Acceptor.

1. Anticodon of tRNA.

Anticodon is a group of three nucleotides in a single-stranded region of each tRNA. **The first two positions in the anticodon must form Watson-Crick base-pairs with the codon, the last position (5' nucleotide) does not have to assume normal Watson-Crick base pairing. Instead this position, the Wobble position, can assume a variety of novel base pairing interactions.**

Anticodon nucleotides interact with codons of mRNA through base-pairing.

Table 29·1 Predicted base pairing between the 5' (wobble) position of the anticodon and the 3' position of the codon

Nucleotide at 5' (wobble) position of anticodon	Nucleotide at 3' position of codon
C	G
A	U
U	A or G
G	U or C
I*	U, A, or C

*I = Inosinate

Each tRNA has a different anticodon sequence, so each base-pairs to different codons.

Look closely at the polarity (5'-3') within the loop, remember that it must be complementary to the mRNA sequence.

2. Amino Acid Acceptor of tRNA.

A tRNA is specific for a single amino acid.

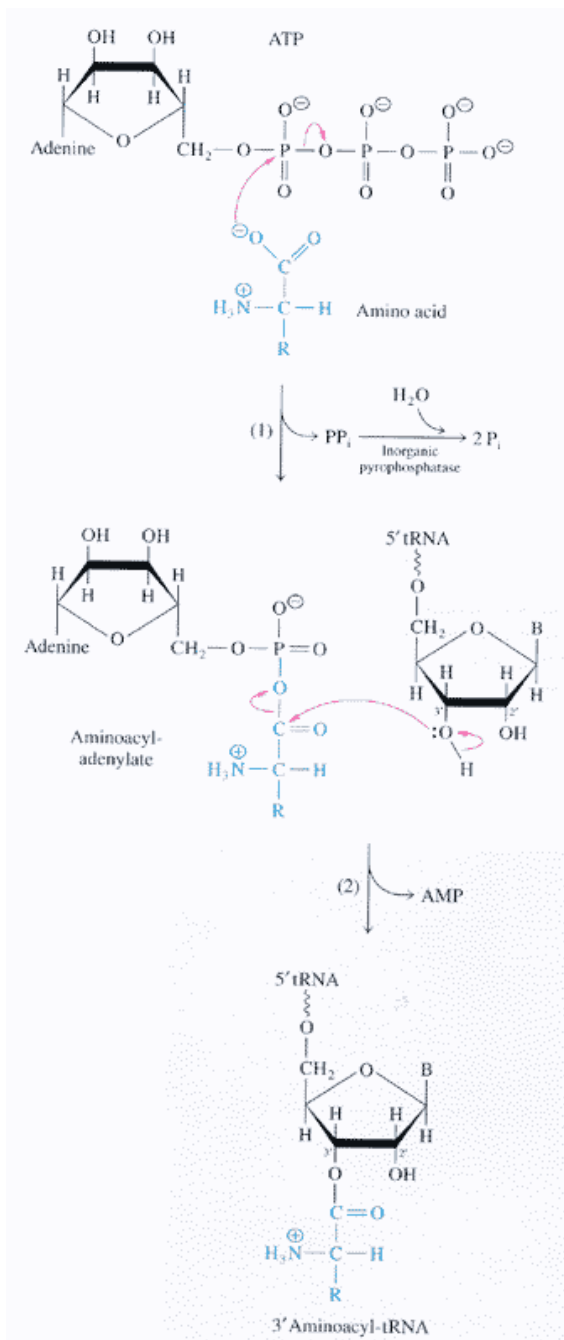
One tRNA for alanine, one for tyrosine, one for valine, etc.

Amino acids are covalently attached to the 3' end of tRNA at the Amino Acid Acceptor.

Amino acids are attached to tRNA by enzymes called **aminoacyl tRNA synthetases.**

Process is called **tRNA Charging.** tRNA is said to be "charged" when an amino acid is attached.

High-energy Phosphate bond of ATP is broken to provide energy for this reaction.



Charging Reaction is very specific.

There is a different **aminoacyl tRNA synthetase** for each amino acid and its corresponding tRNA molecules.

Each synthetase is highly specific both for its amino acid and its tRNA carrier.

Only a tRNA with the correct anticodon is joined with its amino acid.

This specificity is critical for accuracy of translation!

Critical Points:

1. Different tRNAs carry their appropriate amino acid.

Transfer RNAs serve critical role as "Adapters" that translate genetic code into proteins.

2. The anticodon nucleotides of each tRNA have the sequence that is able to basepair and recognize the appropriate mRNA codon for the amino acid the tRNA is carrying.

3. The synthases have **proof reading** activity which allows them to hydrolyze an incorrect pairing, even after it has formed. This proofreading along with other proofing reactions during translation mean the error rate is less than 1/10,000.

Example: methionine codon (5' AUG 3') of mRNA base-paired with anticodon of methionine tRNA (3' UAC 5') and methionine attached at amino acid acceptor.

<u>Amino Acid</u>	<u>Amino-acyl tRNA synthase</u>	<u>tRNA's</u>	<u>Codons</u>
20	20	30-40 prok.	61
		50 eukaryotes	3 stops

Conclusions:

- 1) There is one amino-acyl tRNA synthase per amino acid (they are very specific)
- 2) There is the potential for more than one tRNA per amino acid. Therefore, amino-acyl tRNA synthases must be able to recognize more than one tRNA.
- 3) There is the potential for more than one codon per tRNA. Therefore tRNA's must be able to recognize more than one codon (there is NOT a unique tRNA for each codon).

If perfect Watson-Crick base pairing were required at the codon/anticodon triplet then 61 different tRNA's would be required.

We know that this is not the case, therefore a single tRNA anti-codon must be able to recognize several different mRNA codon triplets.

This greater recognition is possible due to the **wobble basepair interactions** at the third base in the codon/first base in the anti-codon.

Possible wobble codon base pairing (in addition to Watson-Crick):

1. **U-G**
2. **I-C**
3. **I-A**
4. **I-U**

Where U, G, A and C can be in either the codon (mRNA) or the anti-codon (tRNA), and I (inosine) can be found only in the anti-codon.