بسم الله الرحمن الرحيم

Basic Principles and Perspectives in Medical Chemistry and Biochemistry

Nucleic Acids

Part 2

Protein Biosynthesis

5th Medical and Biochemistry (BIQC-101) Lecture

Second Semester

by

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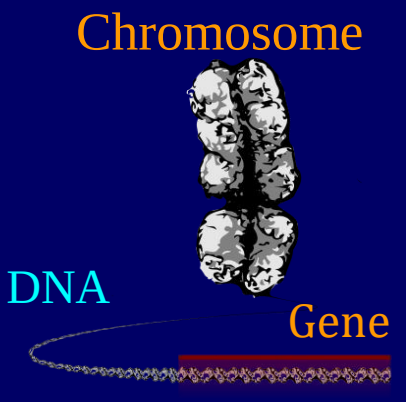
Protein Biosynthesis

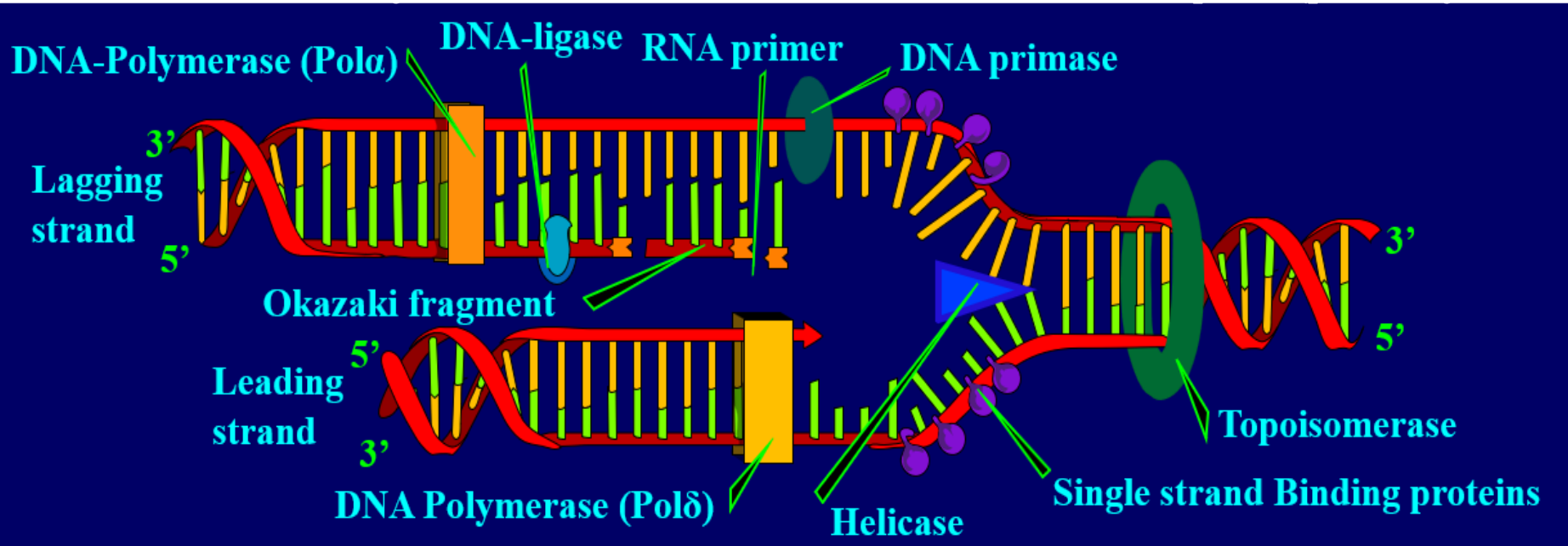
Learning Objectives

1. Comprehend the universal nature of the gene.
2. Be able to define replication of DNA.
3. Know the roles of mRNA, ribosomes, tRNA and amino acids in the process of translation.
4. Understand what start codons and stop codons are.
5. Understand how a polypeptide is built, one amino acid at a time, in the different docking sites of the ribosome.
6. Understand how tRNAs are 'charged' with amino acids.
7. Know that ribosomes consist of a large and a small subunit.
8. Be able to define polysome.

Biological functions

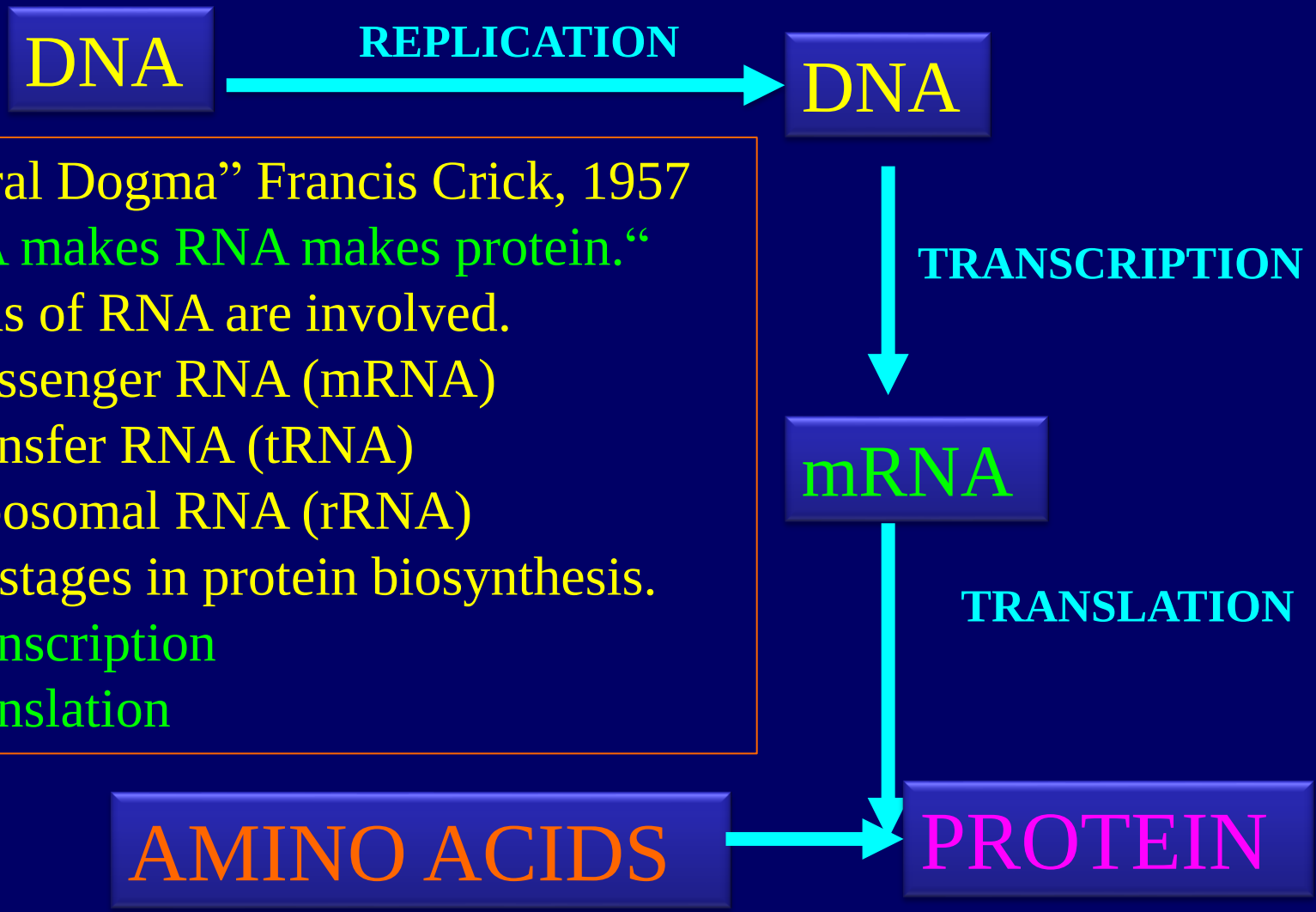


- A gene is a region of DNA (sequence of nucleotides in DNA) that encodes function (carrying the genetic information) . A chromosome consists of a long strand of DNA containing many genes. A human chromosome can have up to 500 million base pairs of DNA with thousands of genes.
- The information carried by DNA is held in the sequence of DNA. So, transmission of genetic information is achieved via complementary base pairing for example:-
- In **transcription**, the DNA sequence is copied into a complementary RNA sequence through the attraction between the DNA and the correct RNA nucleotides, this RNA (mRNA) copy is then used to make a matching protein sequence in a process called translation in protein biosynthesis
- In alternative fashion, a cell may simply copy its genetic information in a process called DNA replication (happened during cell division).



DNA replication: The double helix is unwound by a **helicase** and **topoisomerase.** Each open DNA section, called a **replication fork.** Next, one **DNA polymerase** catalyzes the formation of 5'-3'ester bonds of the leading strand. Another DNA polymerase binds to the lagging strand which grows in the 3'-5' direction. This enzyme makes discontinuous segments (called **Okazaki fragments**) before DNA ligase joins them together. The process is called **semi-conservative replication**, The energy for the synthesis comes from hydrolysis of phosphate groups.

Protein Biosynthesis



"The Central Dogma" Francis Crick, 1957

"DNA makes RNA makes protein."

Three kinds of RNA are involved.

1. Messenger RNA (mRNA)
2. Transfer RNA (tRNA)
3. Ribosomal RNA (rRNA)

Two main stages in protein biosynthesis.

1. Transcription
2. Translation

Ribonucleic acid RNA

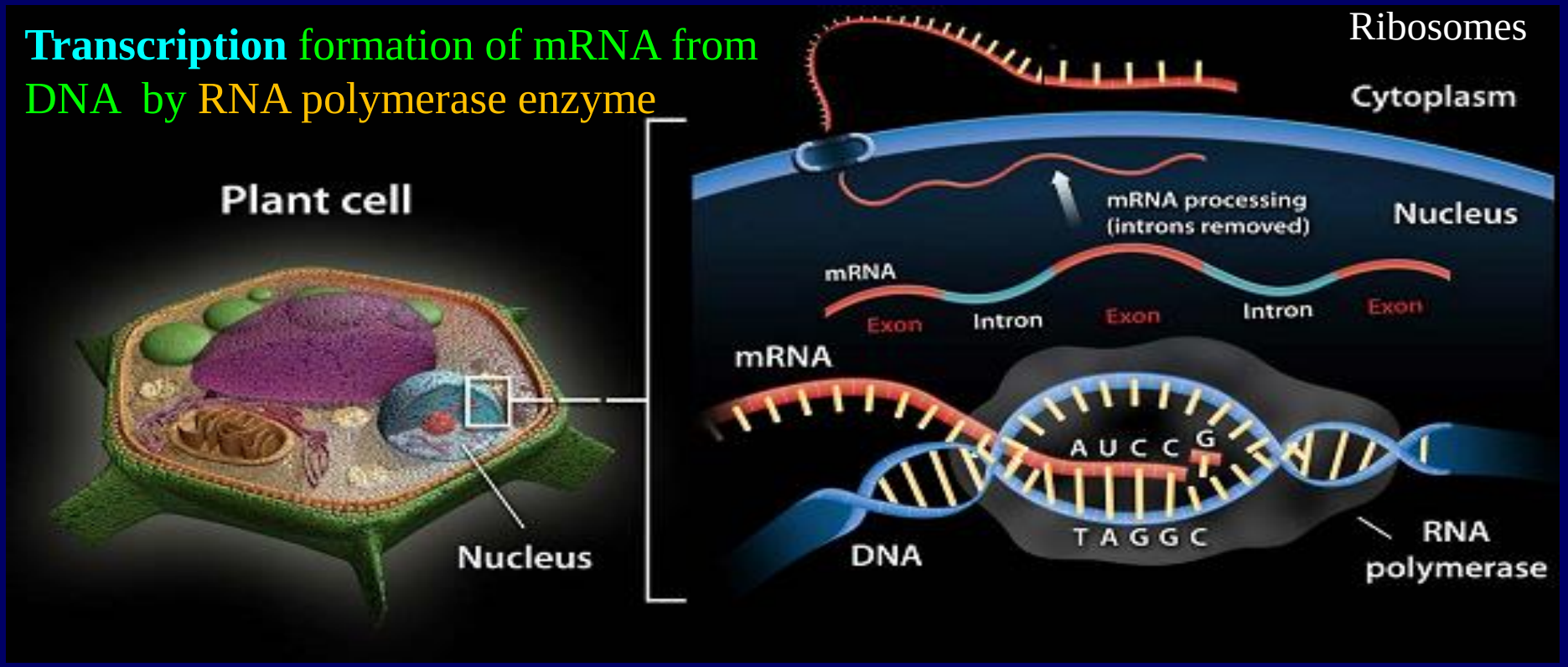
- Ribonucleic acid (RNA) is formed by condensation of nucleotides.
- RNA is a long, **unbranched** macromolecule
- May contain 70 to several thousand nucleotides.
- RNA molecule is usually single stranded.
- RNA contains adenine (A), guanine (G), cytosine (C) and uracil (U).
- Different types of RNA exist in the cell:

1. Messenger RNA (mRNA).
2. Transfer RNA (tRNA).
3. Ribosomal RNA (rRNA).

Messenger (mRNA)

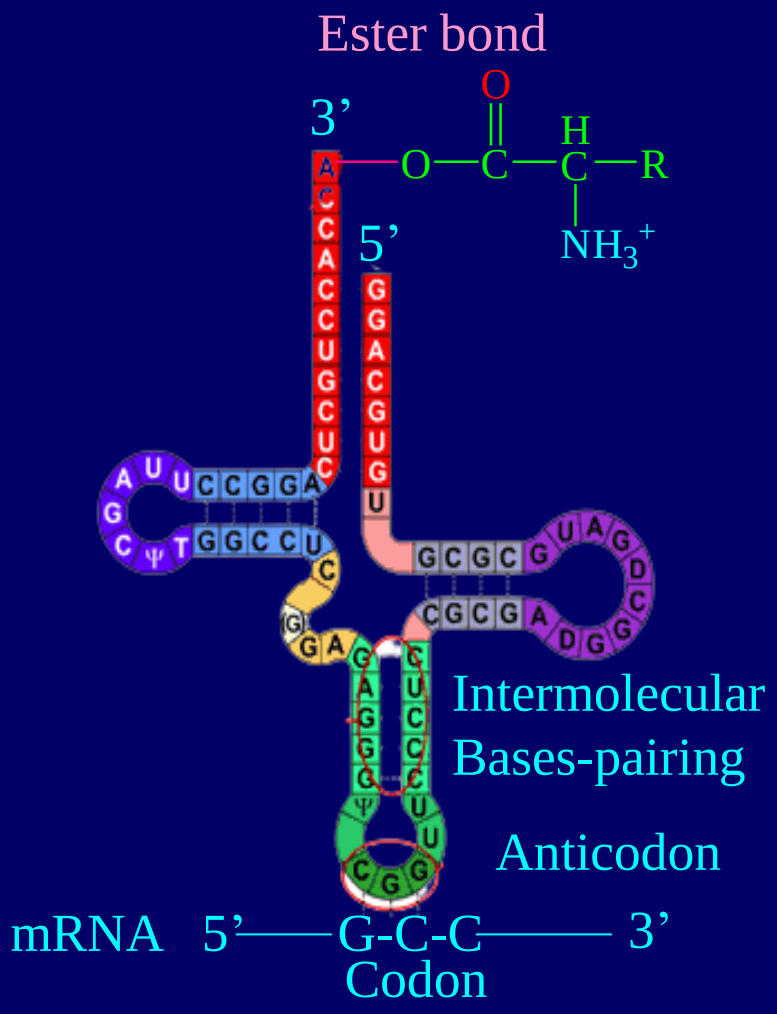
Molecule in cells that carries genetic codes from the DNA in the nucleus to the sites of protein synthesis in the cytoplasm (the ribosomes). Transcription is the first step in gene expression. It involves copying a gene's DNA sequence to make an RNA molecule (mRNA).

Transcription formation of mRNA from DNA by RNA polymerase enzyme



Transfer (tRNA)

- Translates the genetic code from the messenger RNA and brings specific amino acids to the ribosome for protein synthesis
- Each amino acid is recognized by one or more specific tRNA
- tRNA has a tertiary structure that is L-shaped
- One end attaches to the amino acid and the other binds to the mRNA by a 3-base complimentary sequence.

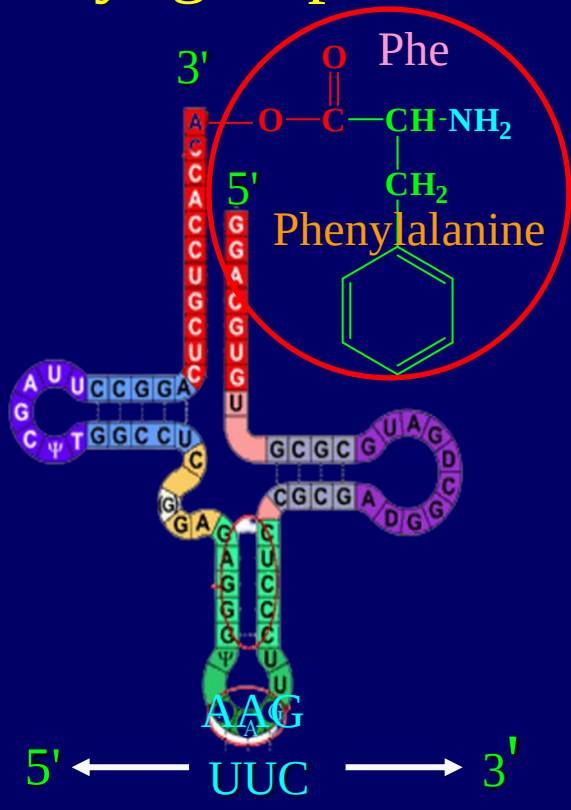


- There are 20 different tRNAs, one for each amino acid.
- Each tRNA consists of sets of three bases (triplet) at its 3' end called anticodon.
- A particular amino acid is attached to the tRNA by an ester linkage involving the carboxyl group of the amino acid and the 3' oxygen of the tRNA hydroxyl group.
- Example—Phenylalanine transfer RNA
- One of the mRNA codons for phenylalanine is:

5' ⟵ UUC ⟶ 3'

- The complementary sequence in tRNA is called the anticodon.

3' ⟵ AAG ⟶ 5'



- The genetic code consists of 64 triplets of nucleotides. These triplets are called **codons**.
- With three exceptions each codon encodes for one of the 20 amino acids used in the synthesis of proteins.
- Most of the amino acids being encoded by more than one codon.
- The genetic code can be expressed as either RNA codons or DNA codons
- RNA codons occur in messenger RNA (**mRNA**) and are the codons that are actually "read" during the synthesis of polypeptides (the process called **translation**).

	U	C	A	G	
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	UCG Trp	G
C					U C A G
A					U C A G
G					U C A G

First letter

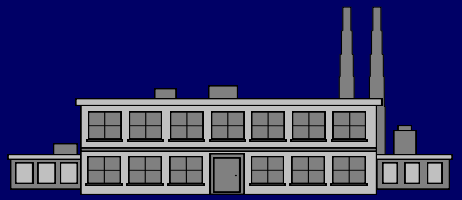
Second letter

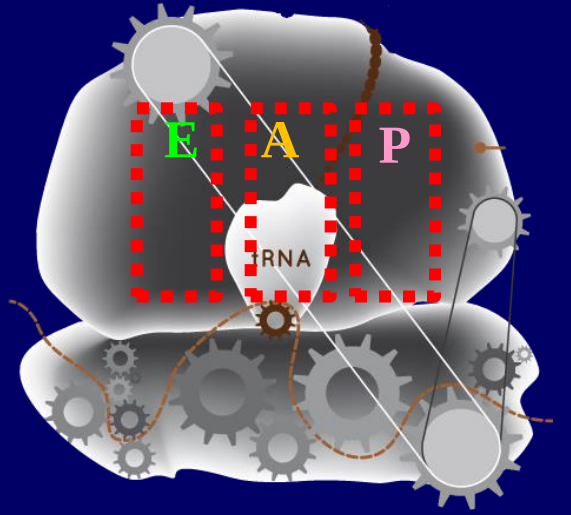
Third letter

	U		C		A		G		
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	UCG	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	CCG	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	ACG	Thr	AAG	Lys	ACG	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	GCG	Gly	G

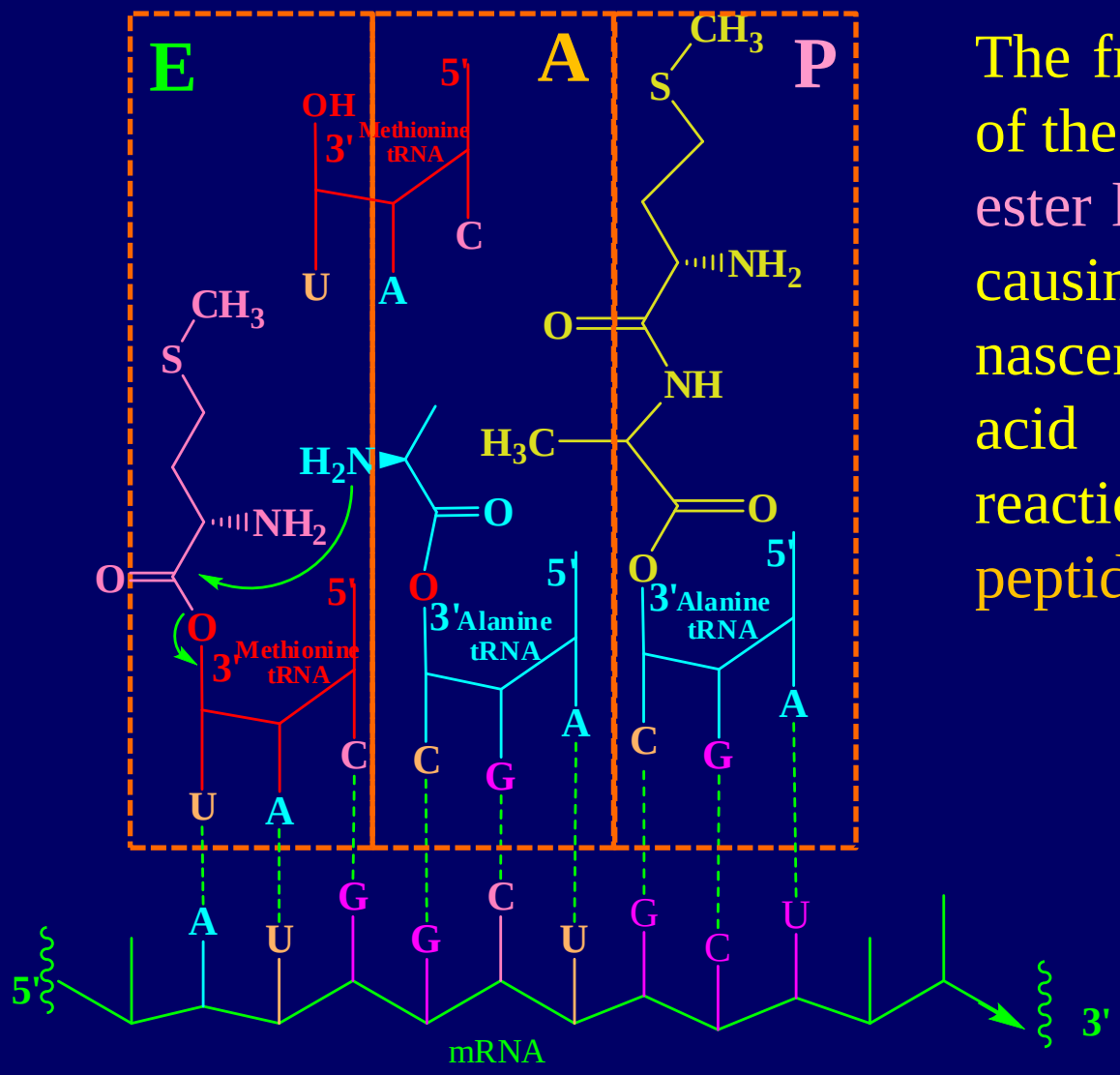
	U	C	A	G	
U	UAA, UGA, and UAG are "stop" codons that signal the end of the polypeptide chain.		UAA Stop UAG Stop	UGA Stop	U C A G
C					U C A G
A	AUU Ile AUC Ile AUA Ile AUG Met	ACU Thr ACC Thr ACA Thr ACG Thr	AAU Asn AAC Asn AAA Lys AAG Lys	AGU Ser AGC Ser AGA Arg ACG Arg	U C A G
G	AUG is the "start" codon. Biosynthesis of all proteins begins with methionine as the first amino acid. This methionine is eventually removed after protein synthesis is complete.				U C A G

Ribosomal (rRNA)





- Found in ribosomes and essential for protein synthesis (protein factory).
- Consist of ribosomal DNA (65%) and proteins (35%).
- Have two subunits, three binding sites called the A, P and E sites:
- A (aminoacyl) site contains an aminoacyl-tRNA (a tRNA esterified to an amino acid on the 3' end).
- P (peptidyl) site contains a tRNA esterified to the nascent peptide.
- E (exit) site contains a tRNA that has been discharged.
- A single mRNA can be translated simultaneously by multiple ribosomes. This is called a polysome.



The free amino (NH2) group of the A site tRNA attacks the ester linkage of E site tRNA, causing transfer of the nascent peptide to the amino acid in the P site. This reaction is takes place in the peptidyl transferase center.

- **Protein biosynthesis** is a core biological process, occurring inside cells, balancing the loss of cellular proteins through the production of new proteins.
- Proteins perform a number of critical functions as enzymes , structural proteins or hormones.
- Protein synthesis can be divided broadly into two phases transcription and translation.

Transcription

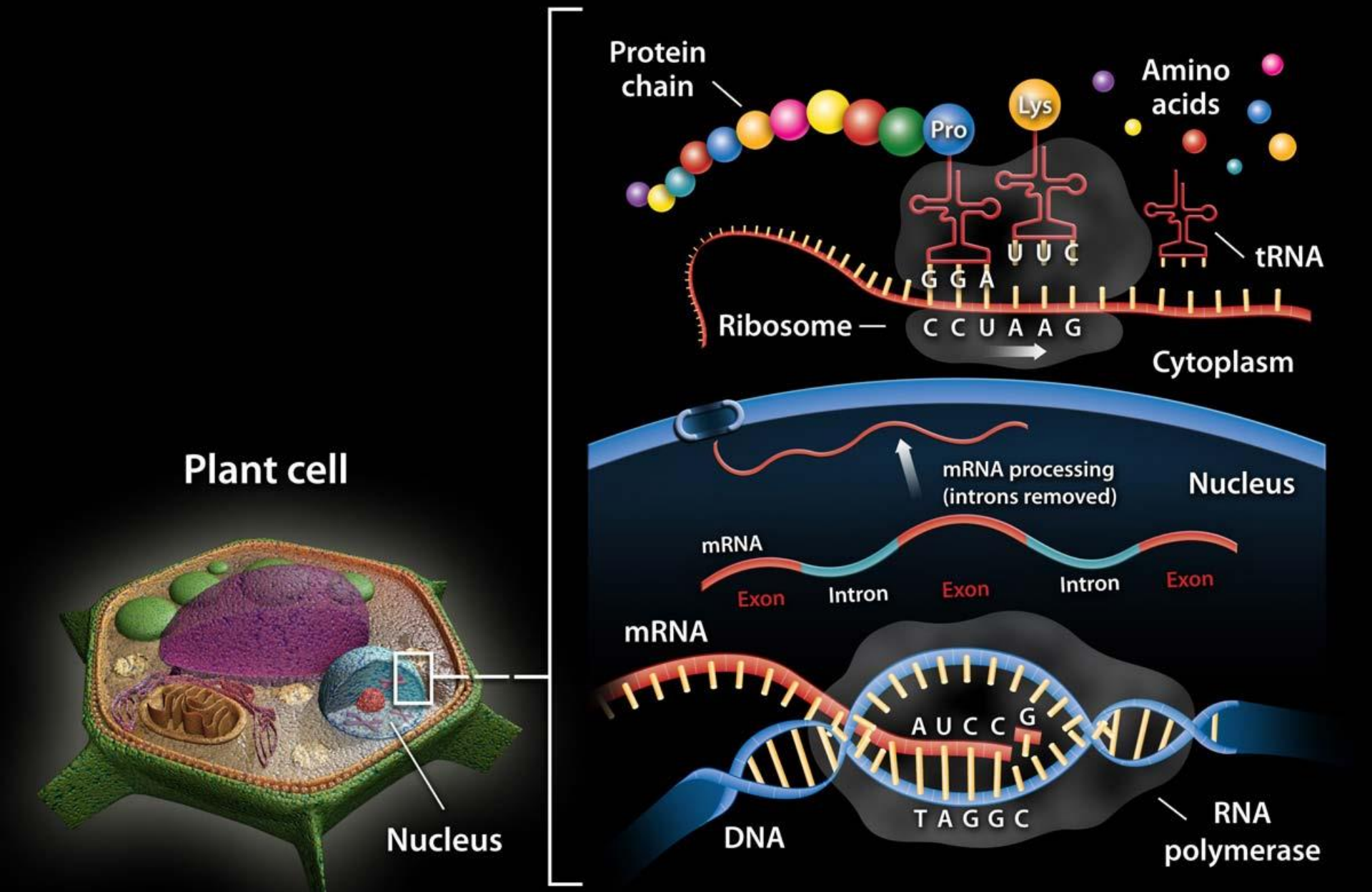
During transcription, a section of DNA encoding a protein, known as a gene, is converted into a template molecule called messenger RNA (mRNA). This conversion is carried out by enzymes, known as RNA polymerases, in the nucleus of the cell.

Translation

- During translation, ribosomes synthesize polypeptide chains from mRNA template molecules.
- In eukaryotes, translation occurs in the cytoplasm of the cell, where the ribosomes are located.
- The ribosome initially attaches to the mRNA at the start codon (AUG).
- The mRNA nucleotide sequence is read in triplets - three adjacent nucleotides in the mRNA molecule correspond to a *single codon*.
- Each tRNA has an exposed sequence of three nucleotides, known as the *anticodon*, which are complementary in sequence to a specific codon that may be present in mRNA.
- For example, the first codon encountered is the start codon composed of the nucleotides AUG.

- The correct tRNA with the anticodon (complementary 3 nucleotide sequence UAC) binds to the mRNA using the ribosome. This tRNA delivers the correct amino acid corresponding to the mRNA codon, in the case of the start codon, this is the amino acid methionine.
- The next codon (adjacent to the start codon) is then bound by the correct tRNA with complementary anticodon, delivering the next amino acid to ribosome. The ribosome then uses its **peptidyl transferase** enzymatic activity to catalyze the formation of the covalent peptide bond between the two adjacent amino acids.
- The ribosome then moves along the mRNA molecule to the third codon. The ribosome then releases the first tRNA molecule
- The next complementary tRNA with the correct anticodon complementary to the third codon is selected, delivering the next amino acid to the ribosome which is covalently joined to the growing polypeptide chain.

- This process continues with the ribosome moving along the mRNA molecule adding up to 15 amino acids per second to the polypeptide chain. Behind the first ribosome, up to 50 additional ribosomes can bind to the mRNA molecule forming a polysome, this enables simultaneous synthesis of multiple identical polypeptide chains.
- Termination of the growing polypeptide chain occurs when the ribosome encounters a stop codon (UAA, UAG, or UGA) in the mRNA molecule. When this occurs, no tRNA can recognize it and a release factor induces the release of the complete polypeptide chain from the ribosome.


Protein chain
Amino acids
Pro
Lys
tRNA
U U C
G G A
Ribosome
C C U A A G
Cytoplasm
Plant cell
mRNA processing (introns removed)
Nucleus
mRNA
Exon
Intron
Exon
Intron
Exon
mRNA
A U C C G
T A G G C
DNA
RNA polymerase
Nucleus

thank you!