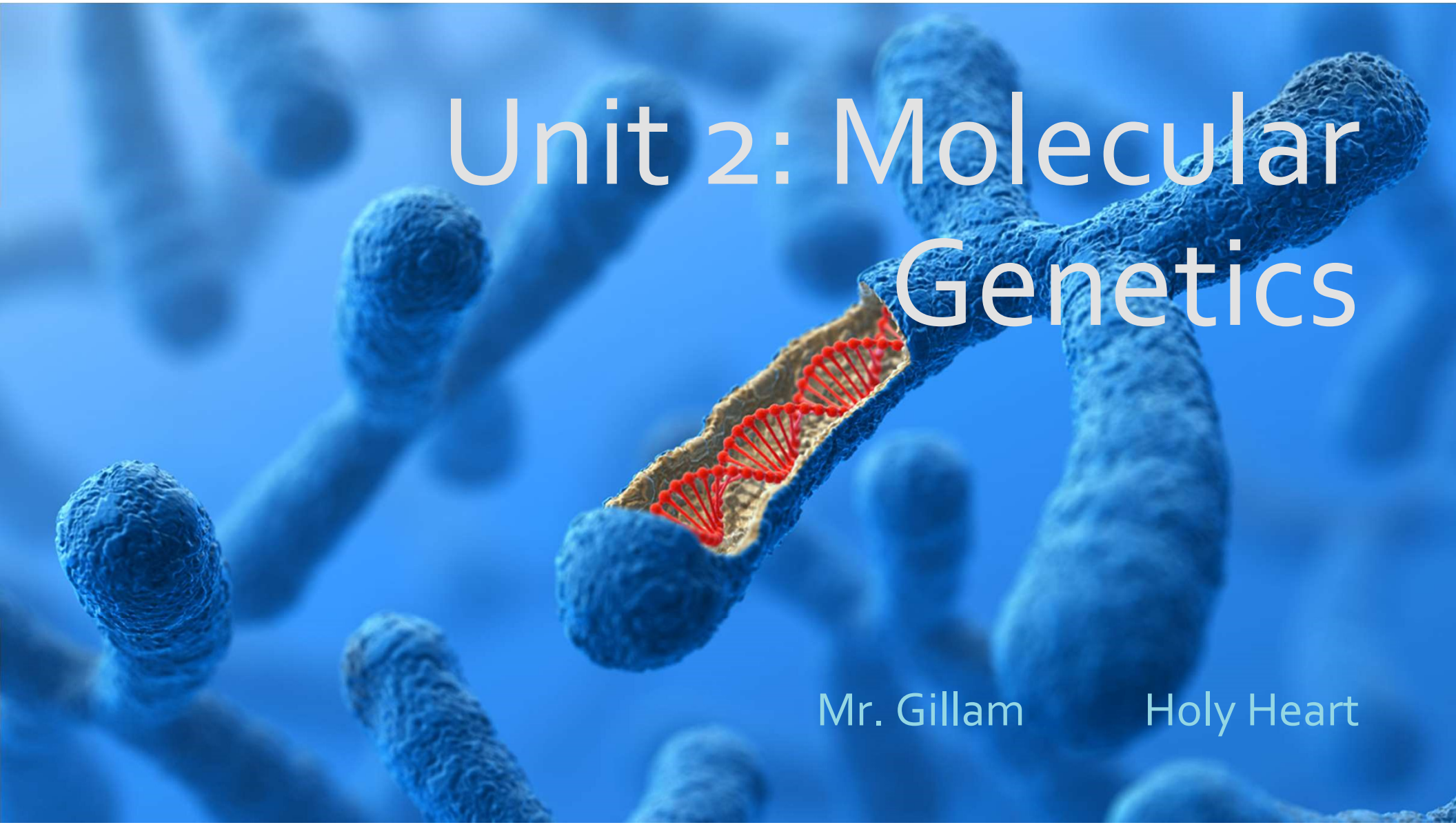


Unit 2: Molecular Genetics

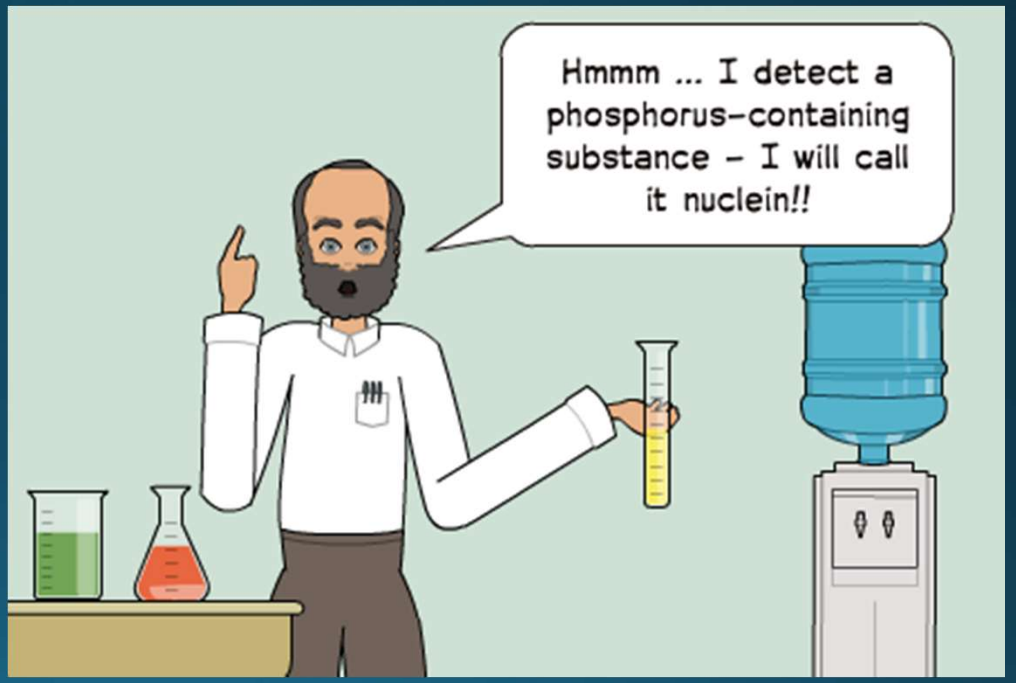
Mr. Gillam Holy Heart

Launch Lab

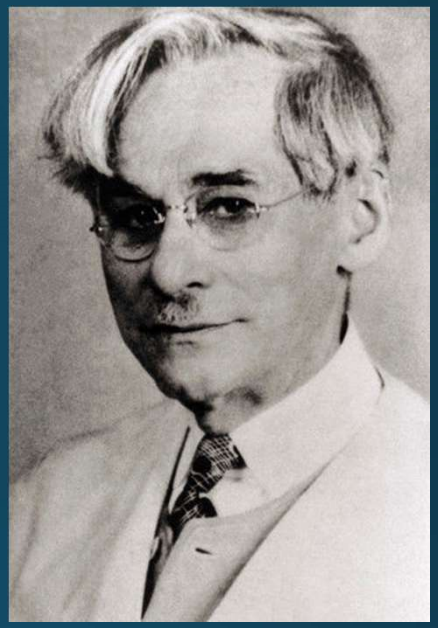
- DNA Extraction Investigation

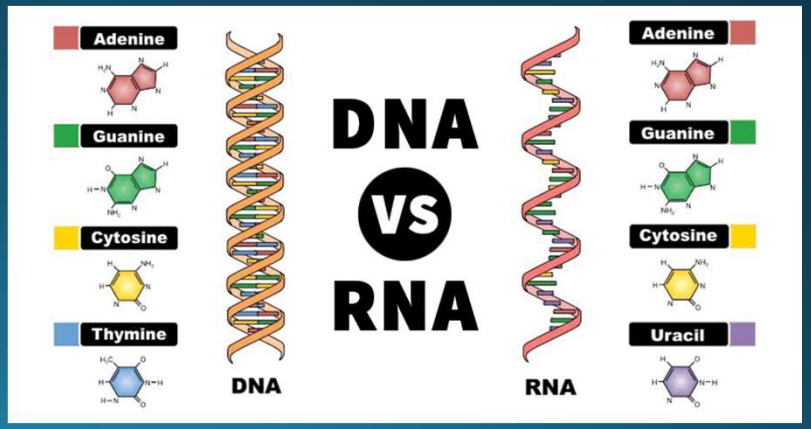
DNA Structure and Replication

- In 1869, a young Swiss physician named **Friedrich Miescher** coined the term "nuclein" to describe a weakly acidic, phosphorus-containing substance that he had isolated from the nuclei of white blood cells. It later became known as **nucleic acid**

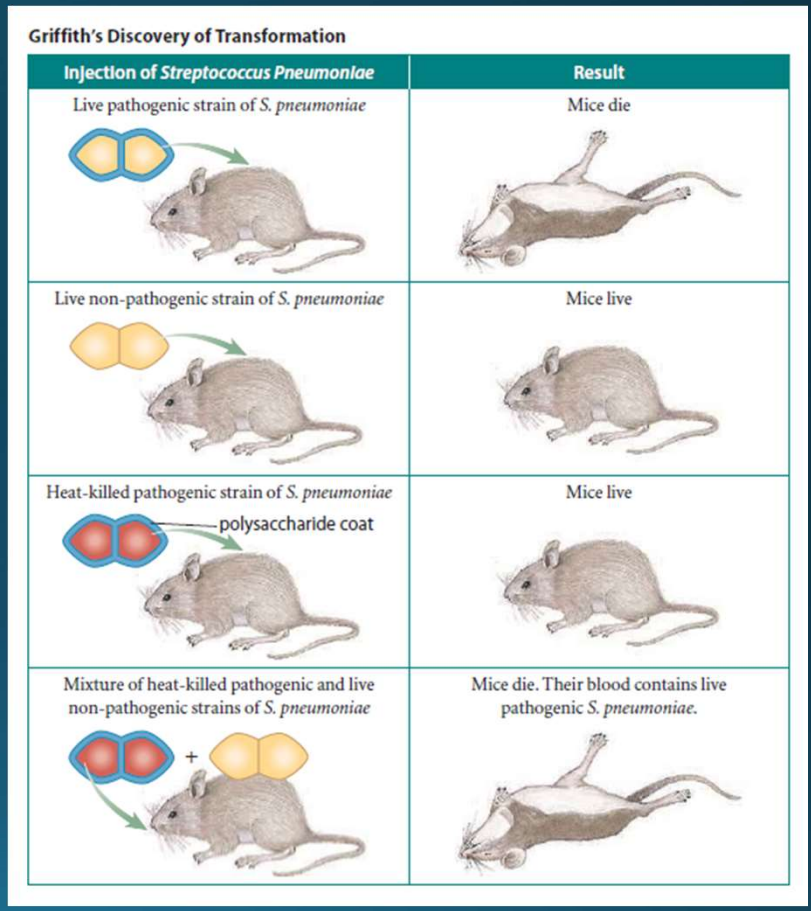


- In the early 1900s, a Russian-born American biochemist named **Phoebus Levene** isolated **two types of nucleic acid**.
- He called them ***ribose nucleic acid (RNA)*** and ***deoxyribose nucleic acid (DNA)*.**
- Levene went on to show that chromosomes are made up of a combination of DNA and proteins.

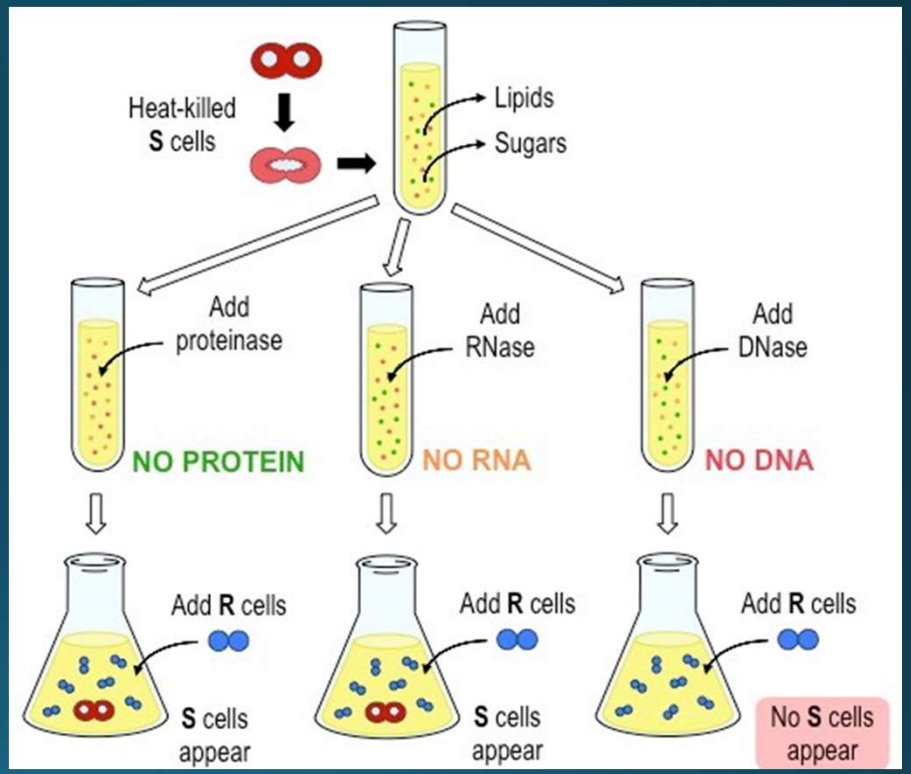




- In 1928, **Frederick Griffith**, an English medical officer, designed an experiment to study the pathogenic (disease causing) bacteria that were responsible for a pneumonia epidemic in London.
- Griffith set up his experiment using dead *Streptococcus pnemoniae* bacteria as a control.
- He discovered that the dead pathogenic bacteria had somehow passed on their disease-causing properties to live, non-pathogenic bacteria Griffith called this phenomenon the ***transforming principle***, because something from the heat-killed pathogenic bacteria must have transformed the living non-pathogenic bacteria to make them disease-causing.



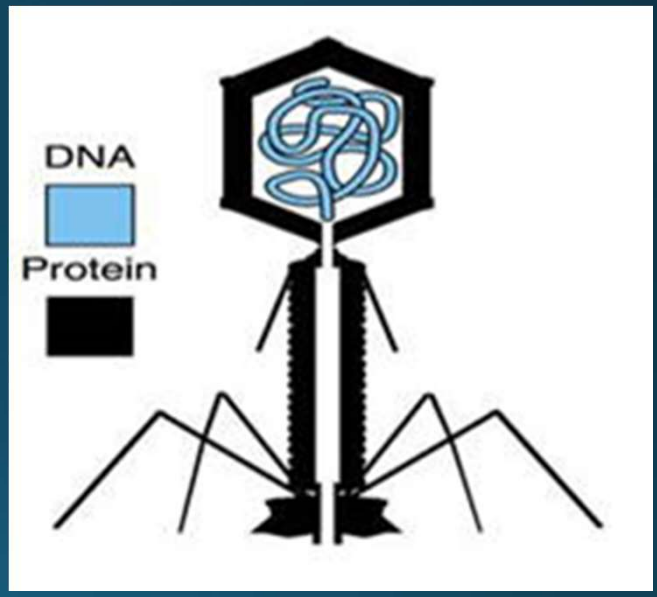
- In 1944, the team of **Oswald Avery, Colin MacLeod, and Maclyn McCarty**, conducted a series of experiments and discovered the following:
- When they treated heat-killed pathogenic bacteria with a protein-destroying enzyme, transformation still occurred.
- • When they treated heat-killed pathogenic bacteria with a DNA destroying enzyme, transformation did not occur.
- **These results provided strong evidence for DNA's role in transformation.** Even so, most scientists still were not prepared to view DNA as the likely source of hereditary material. Instead, they thought that DNA might activate gene-carrying proteins.



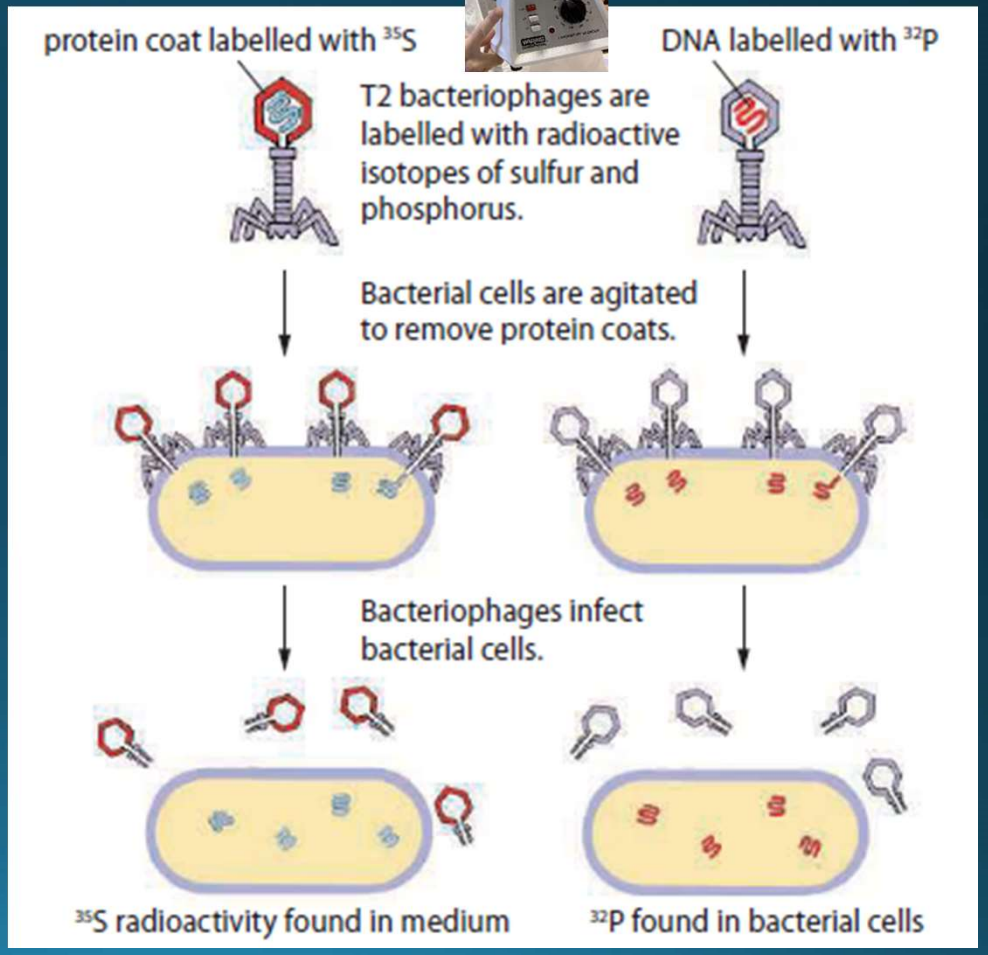
Hershey and Chase: Evidence in Favour of DNA as the Hereditary Material

- Convincing evidence that **DNA, not proteins,** carried genetic information was finally provided in 1952. The American research team of **Alfred Hershey** and **Martha Chase** used a new technology, radioactive labelling, to show that genes are made of DNA.

- Hershey and Chase used a strain of virus known as a **T2 bacteriophage**, which consists of a **protein coat surrounding a length of DNA**.
- This virus attaches to a bacterial cell and injects genetic information into the cell.
- The infected cell manufactures new viruses, and then it bursts.
- The newly released viruses go on to infect other cells.
- To determine whether viral protein or viral DNA was responsible for taking over the genetic machinery of the host cell, Hershey and Chase created **two batches of the virus.**

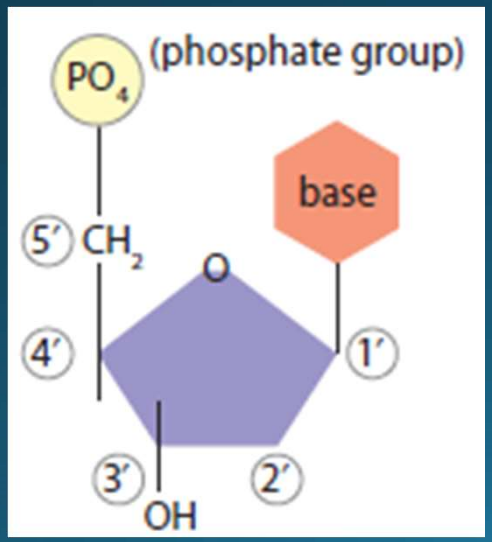


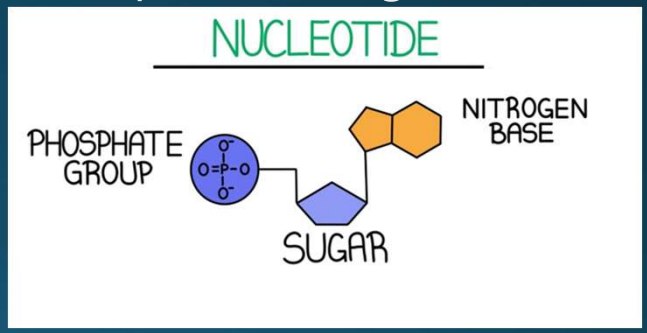
- In one batch, they labelled the **protein coat** using **radioactive sulfur.**
- In the other batch, they labelled the **DNA** with **radioactive phosphorus**.
- The labelled viruses were allowed to infect bacterial cells.
- The cells were then **agitated in a blender** to separate the viral coats from the bacterial cells.
- Each medium was tested for radioactivity.
- **The results demonstrated that viral DNA, not viral protein, enters the bacterial cell.**

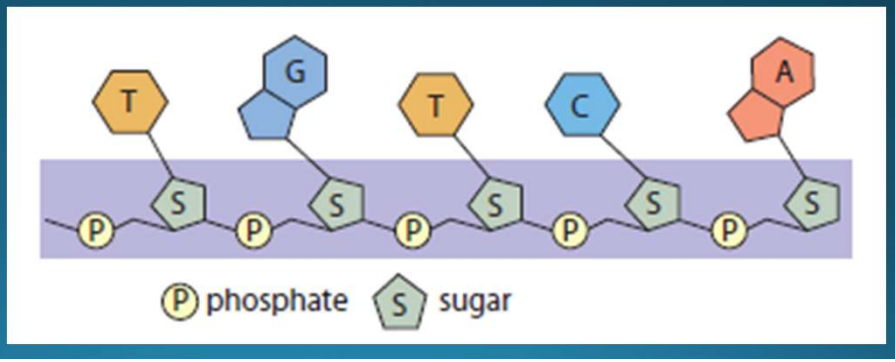


The Structure of DNA and RNA

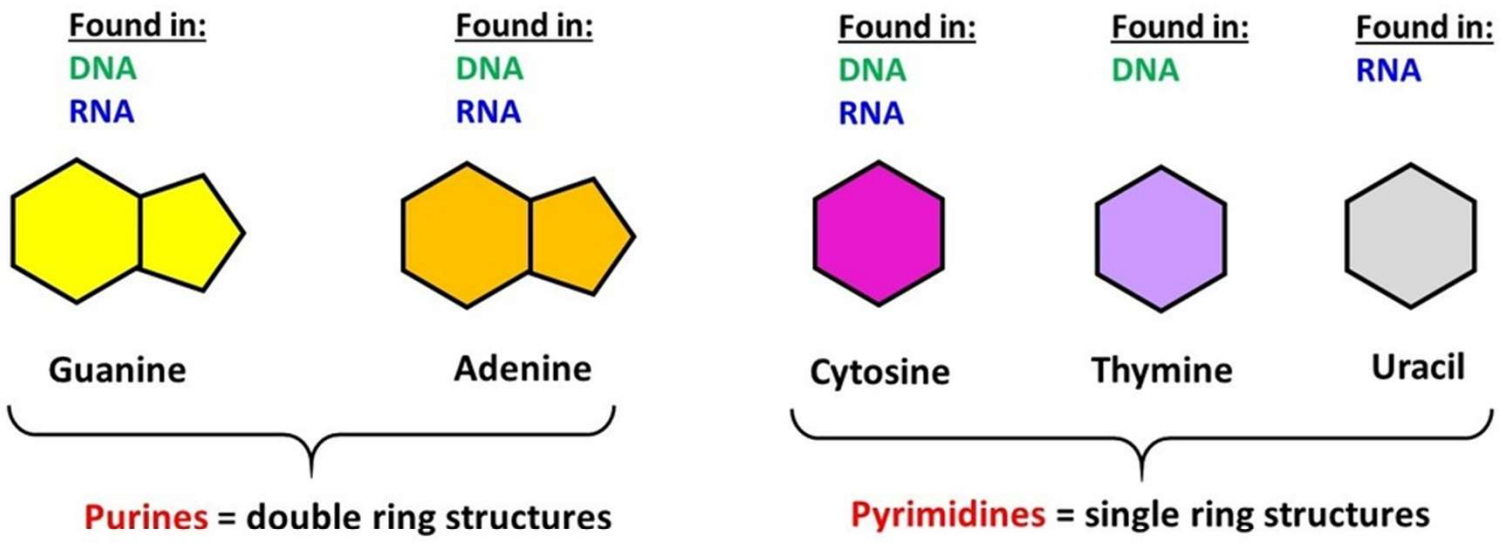
- After isolating DNA and RNA, **Levene** determined that both molecules are made up of long chains of individual units he called nucleotides. Levene also determined that nucleic acids are made up of long chains of nucleotides, strung together
- **nucleotide** repeating unit of nucleic acids; composed of sugar, phosphate, and nitrogenous groups







There are five nitrogenous bases in total:

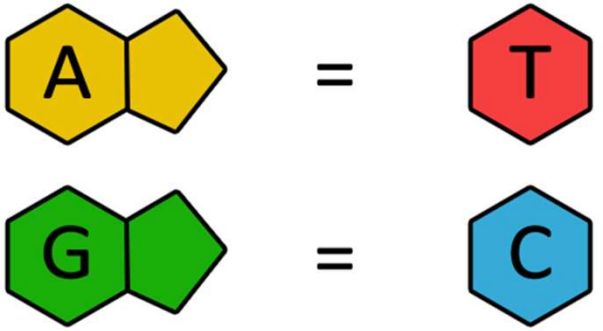


Chargaff's Rule

- **Erwin Chargaff** found that the nucleotides are not present in equal amounts as Levene said.

- **complementary base pairs** refers to hydrogen-bonded base pairs (A-T, C-G)

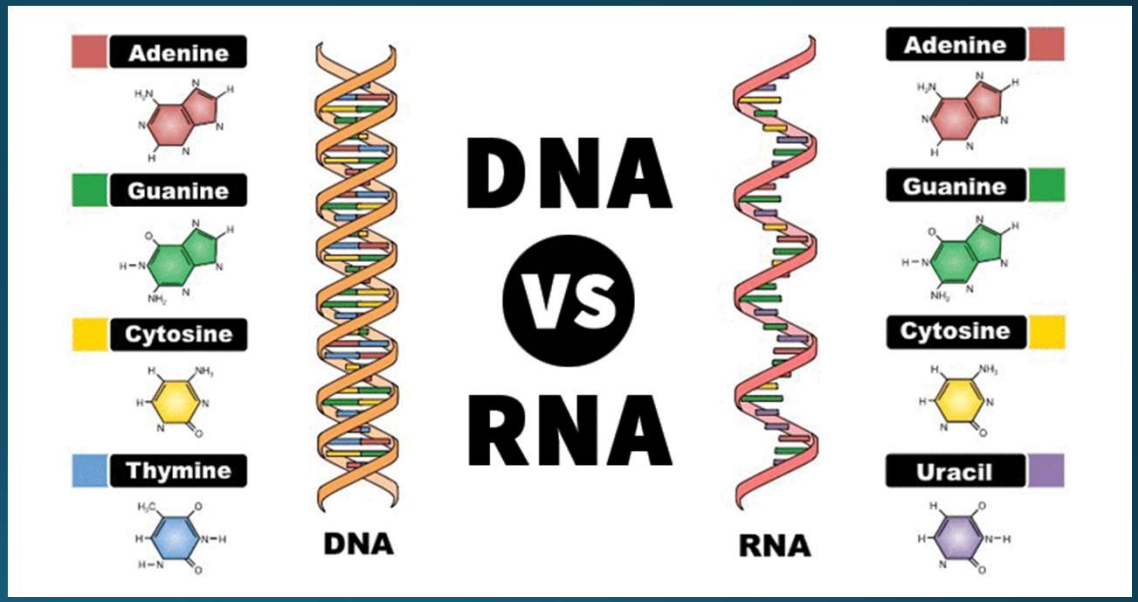
- **Chargaff's rule** in a DNA sample, the amount of adenine is about the same as thymine and the amount of cytosine is about the same as guanine



Organism	A (Adenine)	T (Thymine)	G (Guanine)	C (Cytosine)
Mycobacterium tuberculosis	15.1	14.6	34.9	35.4
Escherichia coli	26.0	23.9	24.9	25.2
Yeast	31.3	32.9	18.7	17.1
Herring	27.8	27.5	22.2	22.6
Rat	28.6	28.4	21.4	21.5
Human	30.9	29.4	19.9	19.8

- The four nitrogenous bases that are found in DNA nucleotides are **adenine (A), guanine (G), cytosine (C), and thymine (T).**

- **RNA** has the base **uracil (U)** instead of **thymine**

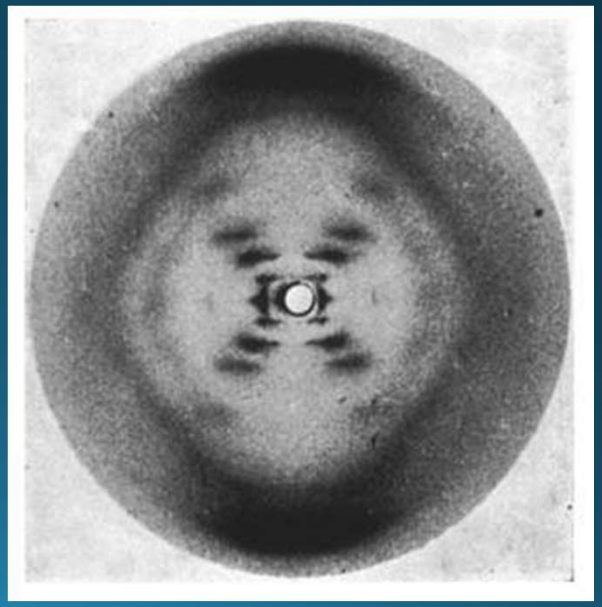


- Activity 15.1 DNA Deductions

- Exit Card #8

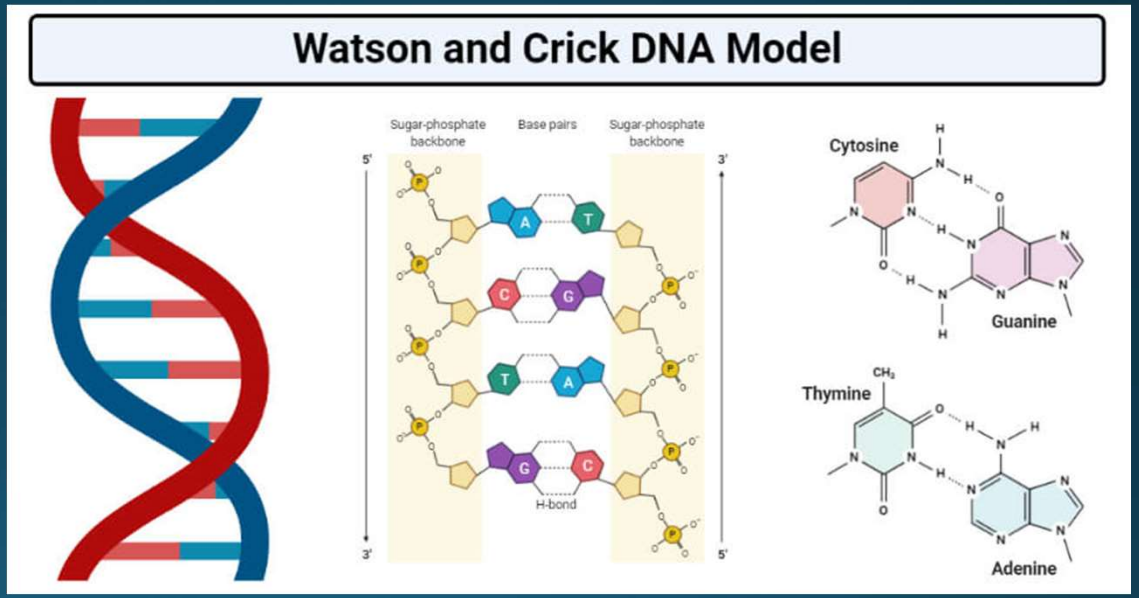
The Three-Dimensional Structure of DNA

- Early in the 1950s, British scientist **Rosalind Franklin** used **X-ray photography** to analyze the structure of DNA.
- Her observations provided crucial new information about the molecular structure of DNA.
- She was able to conclude that DNA has a **helical structure** with two regularly repeating patterns
- From her observations, she concluded that the **nitrogenous bases were located on the inside of the helical structure**, and the **sugar-phosphate backbone was located on the outside**

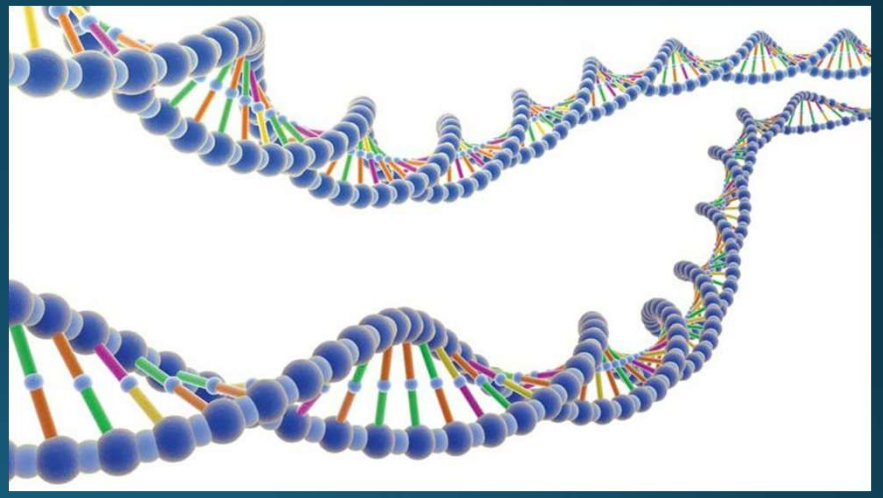


Watson and Crick

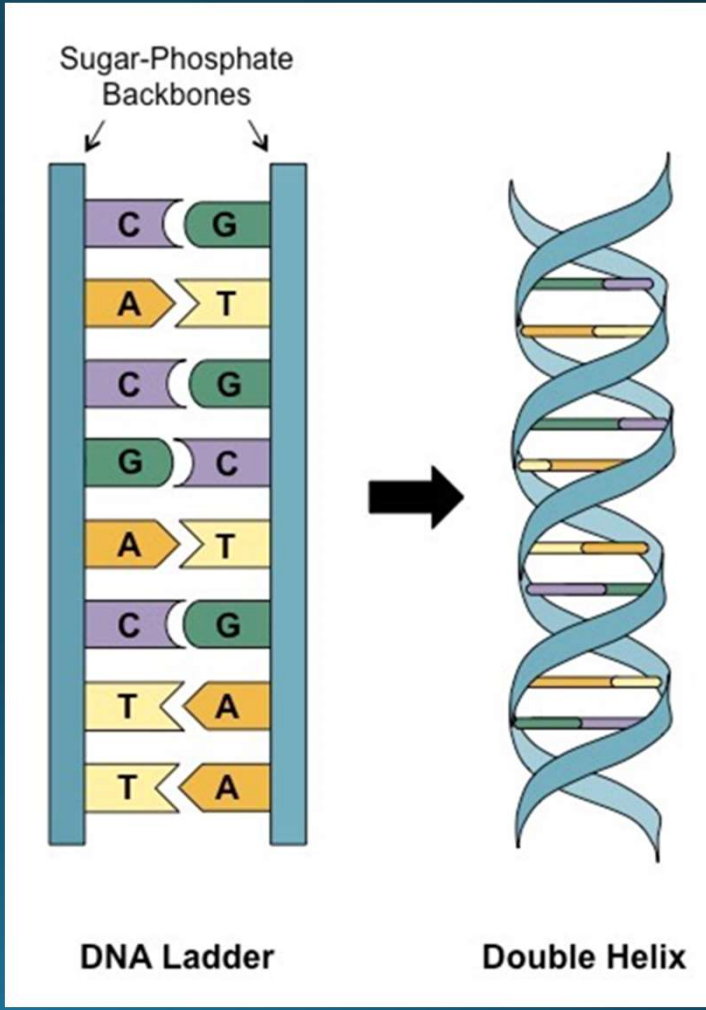
- In 1953, James Watson and Francis Crick published a two-page paper describing a **double helix** model. This model soon became accepted as the molecular structure of DNA.



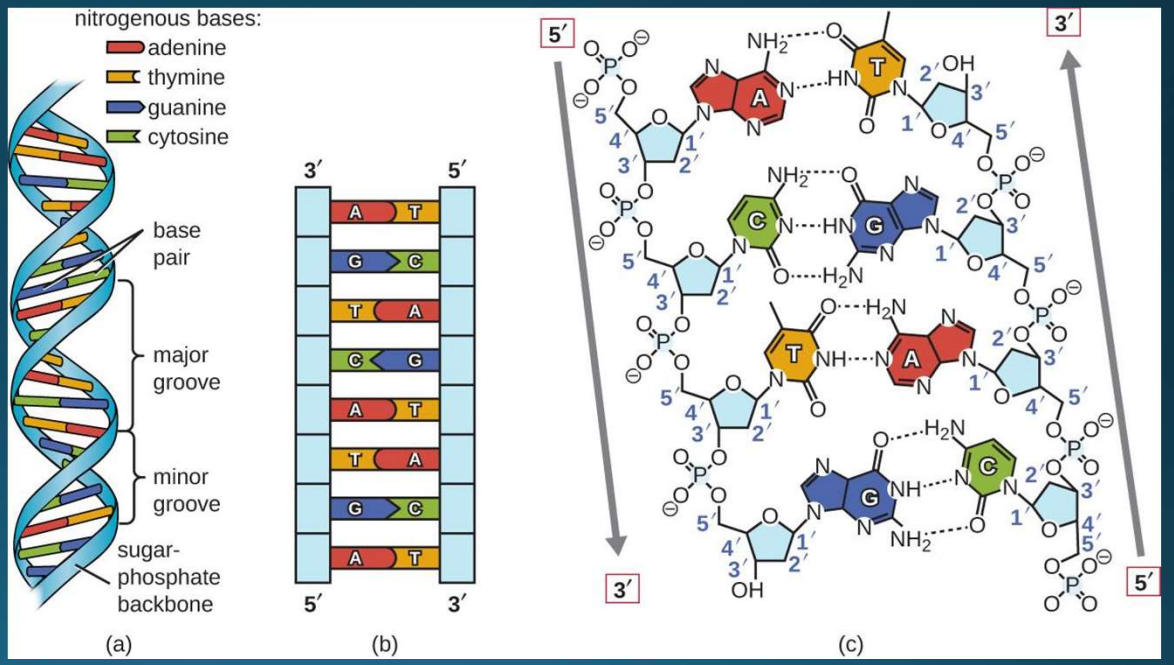
- The discovery of the double helix marked a milestone in the history of science.
- The discovery yielded ground-breaking insights into the genetic code and protein synthesis and helped to produce new and powerful genetic engineering techniques.



- DNA is a thread-like molecule, made up of **two long strands** of nucleotides that are bound together in a spiral shape called a *double helix*. If the helix were unwound, the DNA molecule would look something like a twisted ladder. The "handrails" of the ladder are the **sugar-phosphate backbones** of the two nucleotide strands. The "rungs" are the bases that protrude inward at regular intervals along each strand.

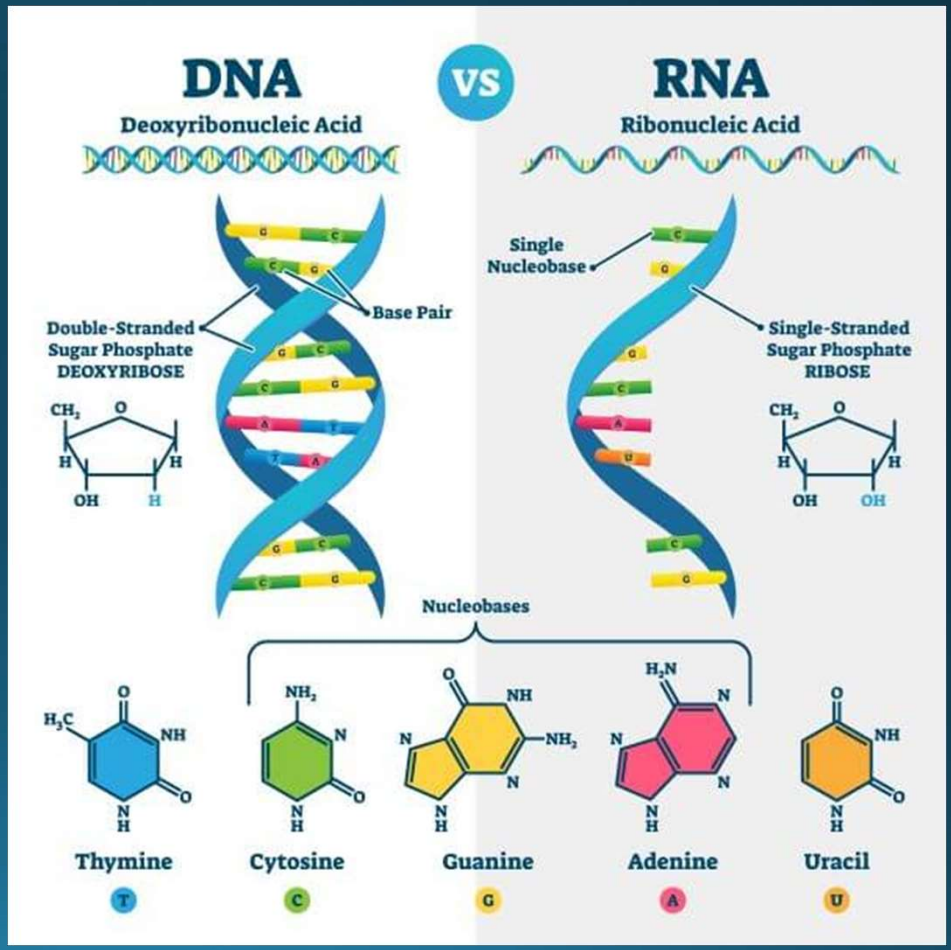


- The two strands are ***antiparallel***, as well. That is, the phosphate bridges run in opposite directions in the two strands. Each end of a double-stranded DNA molecule contains the 5′ end of one strand and the 3′ end of the complementary strand.



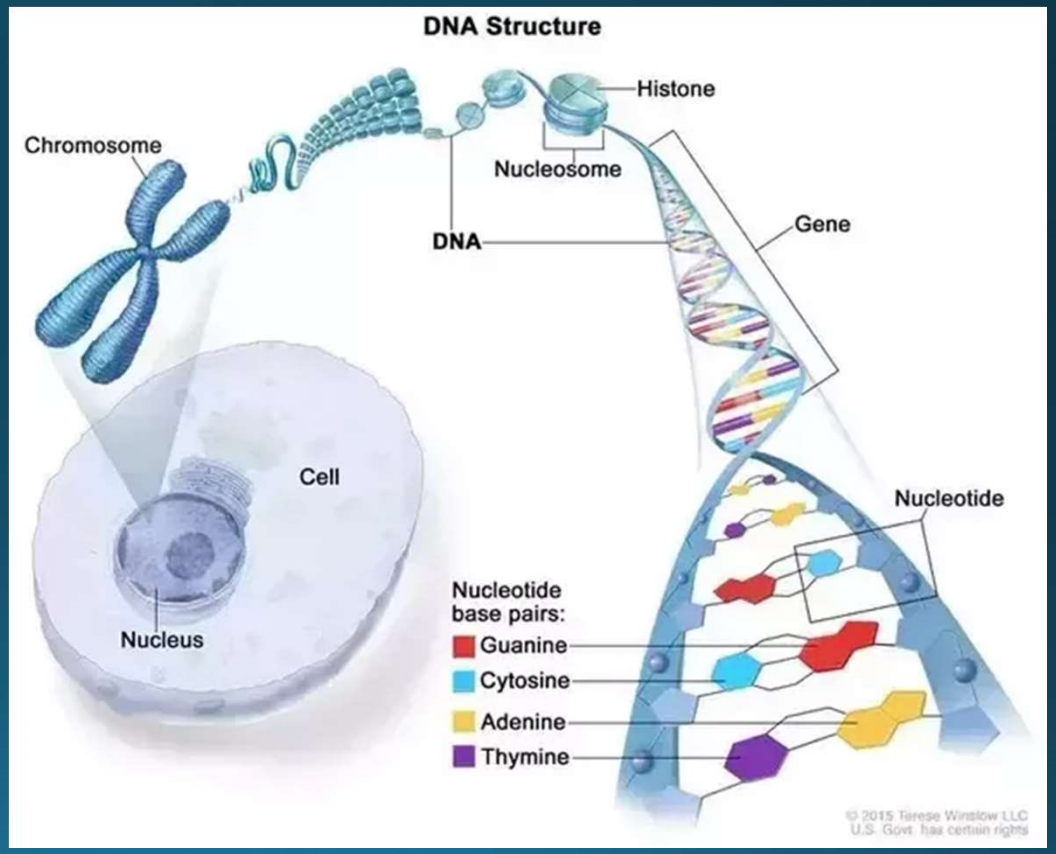
The Structure of RNA vs DNA

- The molecular structure of RNA is similar to the molecular structure of DNA, with three key differences:
- **1.) The sugar component of RNA is ribose rather than deoxyribose.**
- **2.) RNA does not have the nucleotide thymine (T). In its place is the nucleotide uracil (U).**
- **3.) RNA remains single-stranded, although the single strand can sometimes fold back on itself to produce regions of complementary base pairs.**

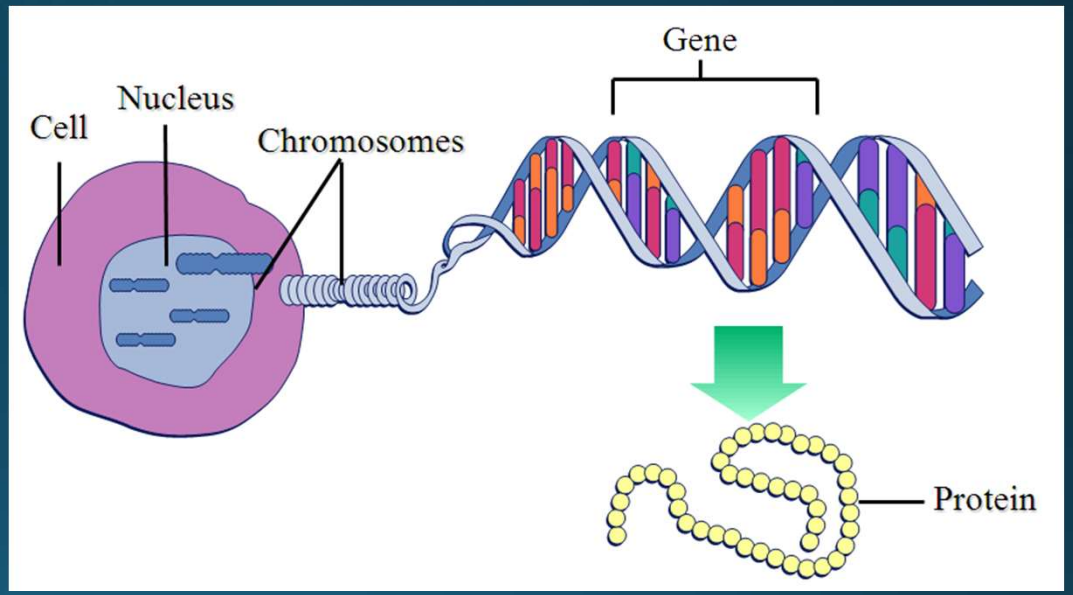


Genes and the Genome

- **chromosome** a length of DNA and associated protein; condensed form of genetic material



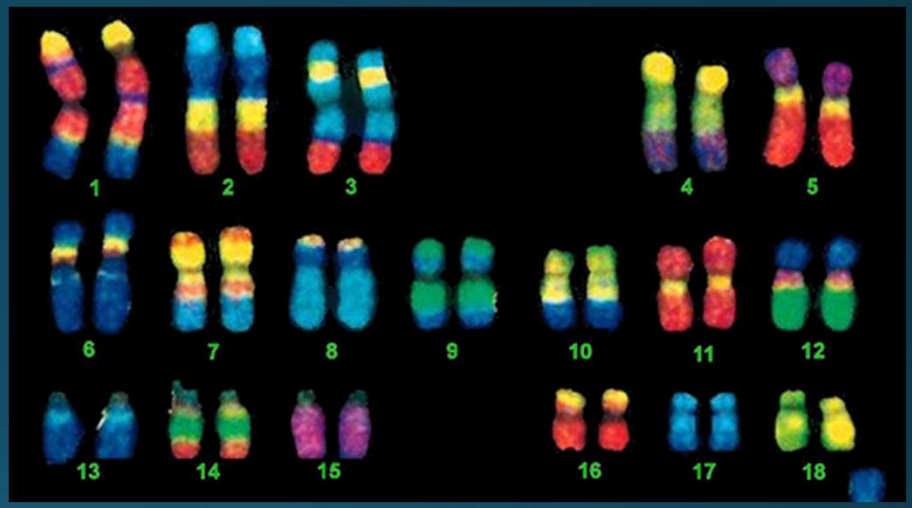
- **gene** a functional sub-unit of DNA that directs the production of one or more polypeptides (protein molecules)
- **genome** the total DNA in an organism's cells



The Human Genome

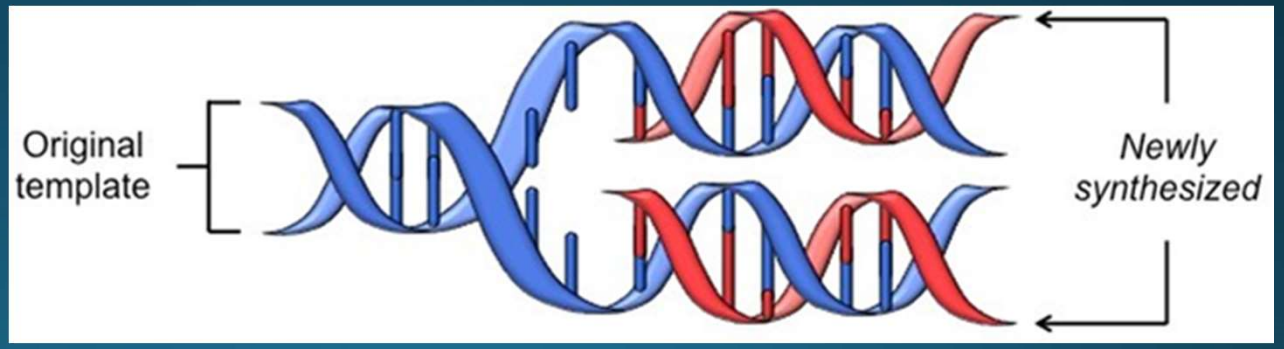
- In humans, chromosome 4 is about 200,000,000 bases long and has about 800 genes, while chromosome 19 is only 55,000,000 bases long but has almost 1500 genes.

- The total human genome is about 3 billion base pairs, and it includes an estimated **20,000 to 25,000 genes.**



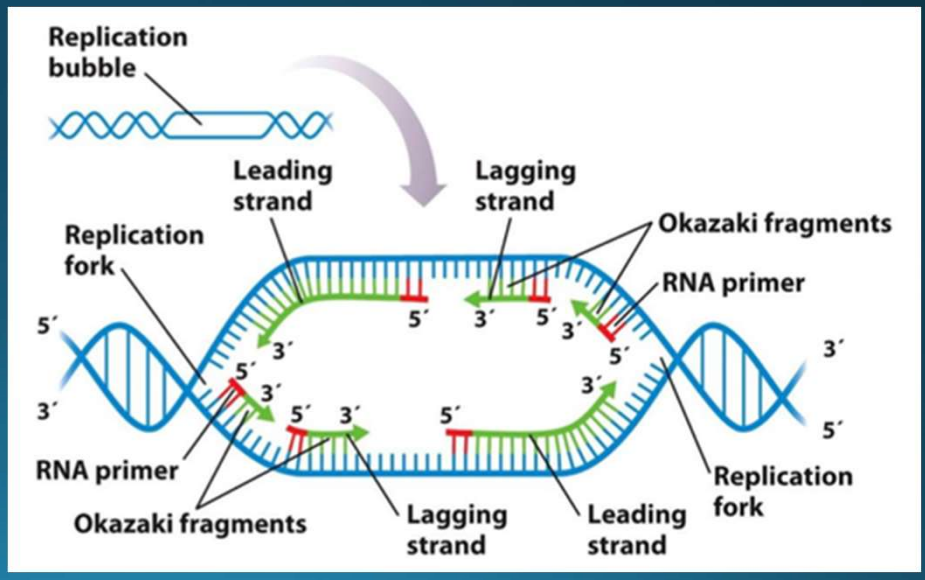
DNA Replication (DNA Synthesis)

- **replication** in genetics, the process of copying DNA
- ***semi-conservative model*** each new molecule of DNA contains one strand of the original complementary DNA molecule and one new parent strand. Thus, each new DNA molecule conserves half of the original molecule.
- Four stages of DNA Replication
- **1. Initiation**
- **2. Elongation**
- **3. Termination**
- **4. Proofreading**



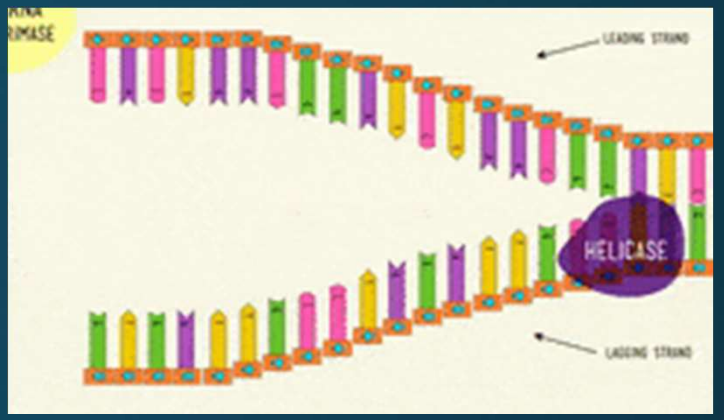
Initiation

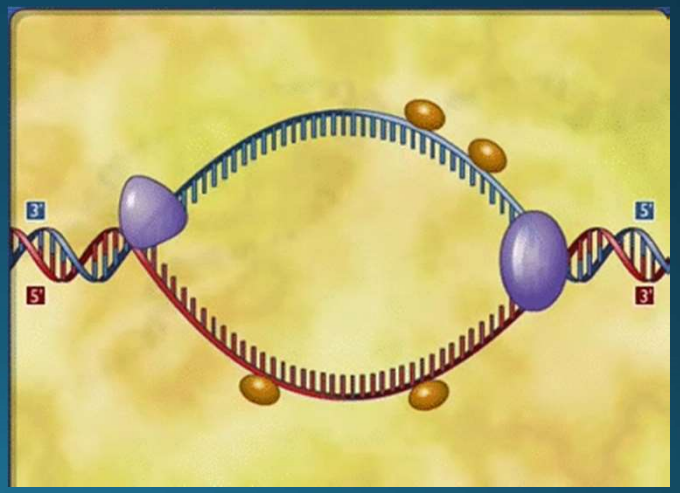
- **replication origin** nucleotide sequence where DNA replication begins
- **helicase** enzyme that bind to the DNA at the replication origin.
- The helicases cleave and unravel a segment of the double helix. This opening up of a region of DNA creates two Y-shaped areas at each end of the unwound area.
- The oval-shaped unwound area is called a **replication bubble**.
- Each Y-shaped end of the bubble is called **replication fork**



Elongation

- **elongation** in replication, the process of joining nucleotides to extend a new strand of DNA
- **DNA polymerase** adds new nucleotides to the 3' OH group of an existing nucleotide strand; dismantles the RNA primer; proofreads base pairing
- **Primase** synthesizes an RNA primer to begin the elongation process
- First, elongation can only take place in the 5′ to 3′ direction.
- Second, a short strand of RNA, known as a ***primer***, must serve as a starting point for the attachment of new nucleotides.
- Replication occurs in a slightly different way along each strand of the parent DNA.





- One strand is replicated continuously in the 5′ to 3′ direction. This strand is known as the leading strand.
- The other strand, known as the lagging strand, is replicated in short segments.

- **leading strand** in replication, the strand made continuously
- **lagging strand** in replication, the strand made in segments

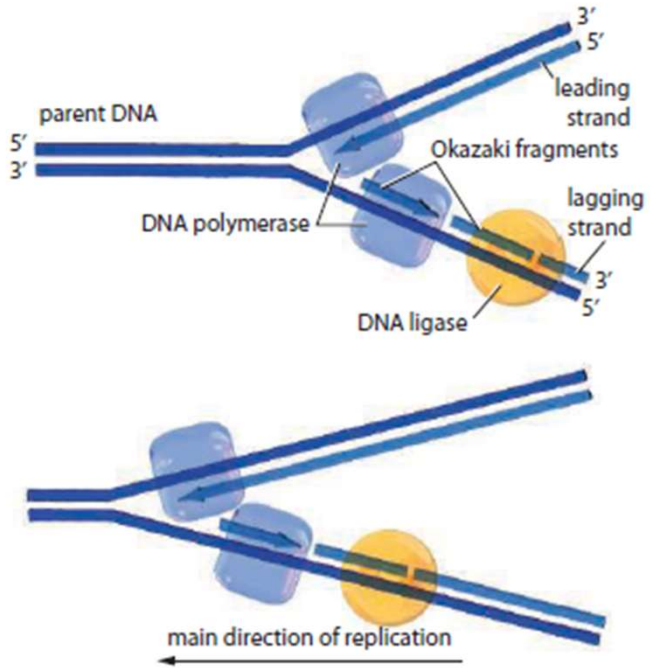


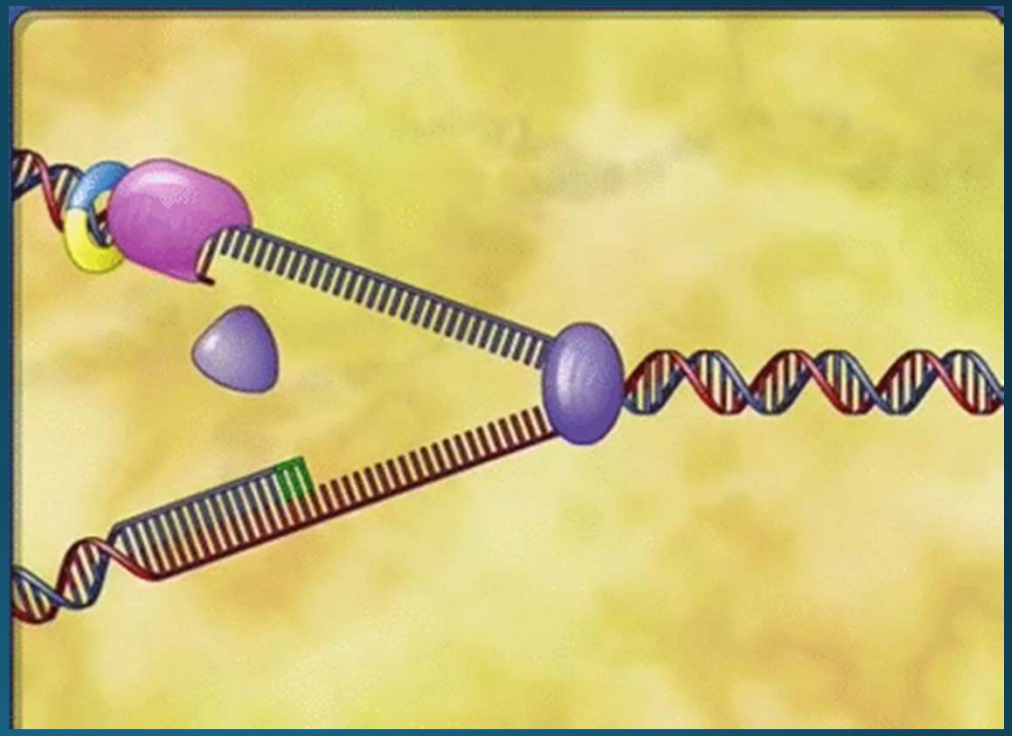
Figure 15.10 During DNA synthesis, the overall direction of elongation is the same along both strands, but elongation occurs differently. On the leading strand, DNA synthesis takes place along the DNA molecule in the same direction as the movement of the replication fork. On the lagging strand, DNA synthesis proceeds in the opposite direction to the movement of the replication fork. As well, the lagging strand is synthesized in short fragments.

Elongation on the Lagging Strand

- Nucleotides are still added in the 5′ to 3′ direction on the lagging strand, but the new DNA is synthesized in short segments called Okazaki fragments.

- **Okazaki fragment** short nucleotide fragments of the lagging strand

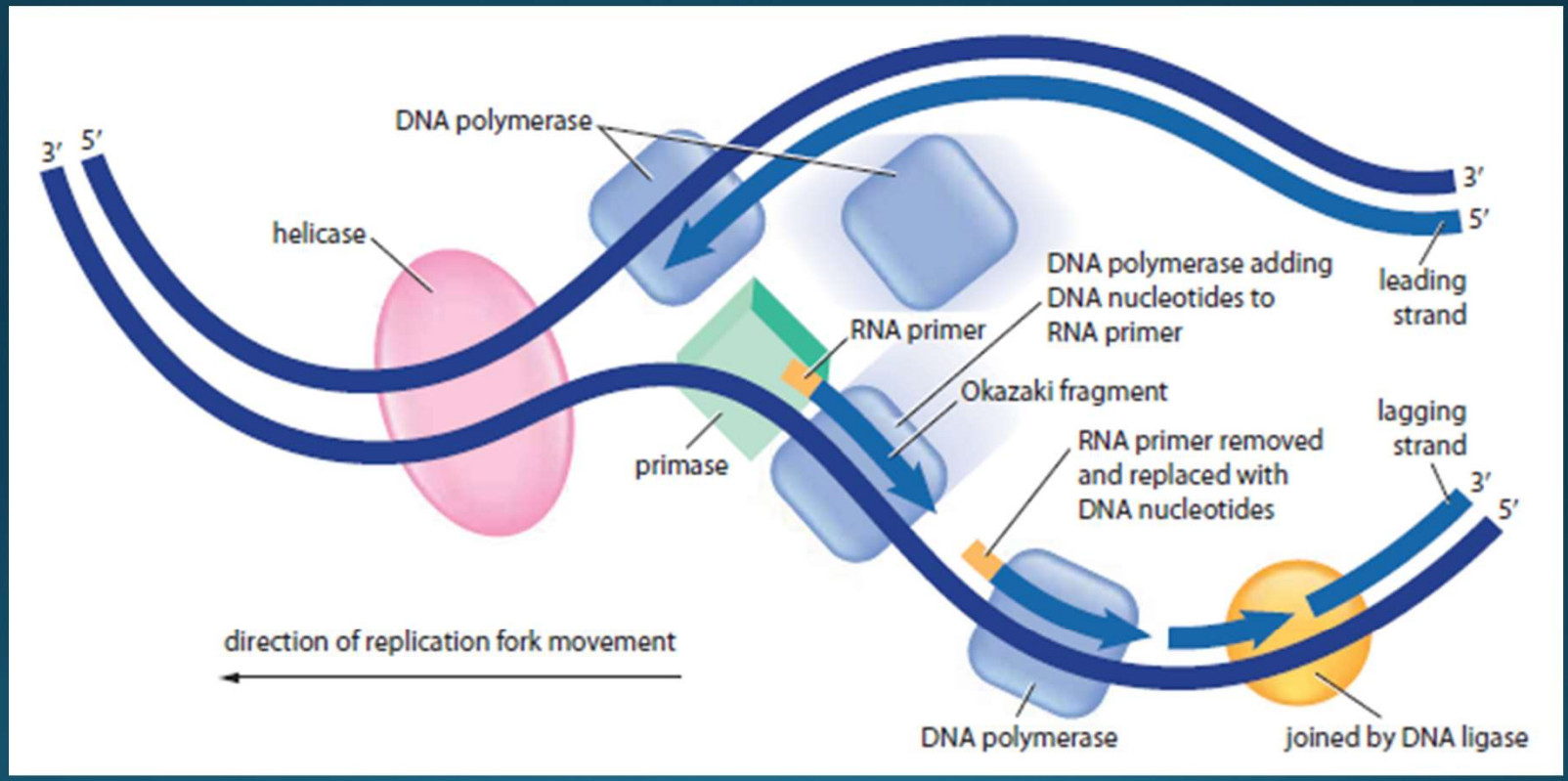
- **DNA ligase** Joins together Okazaki fragments in the lagging strand



Termination

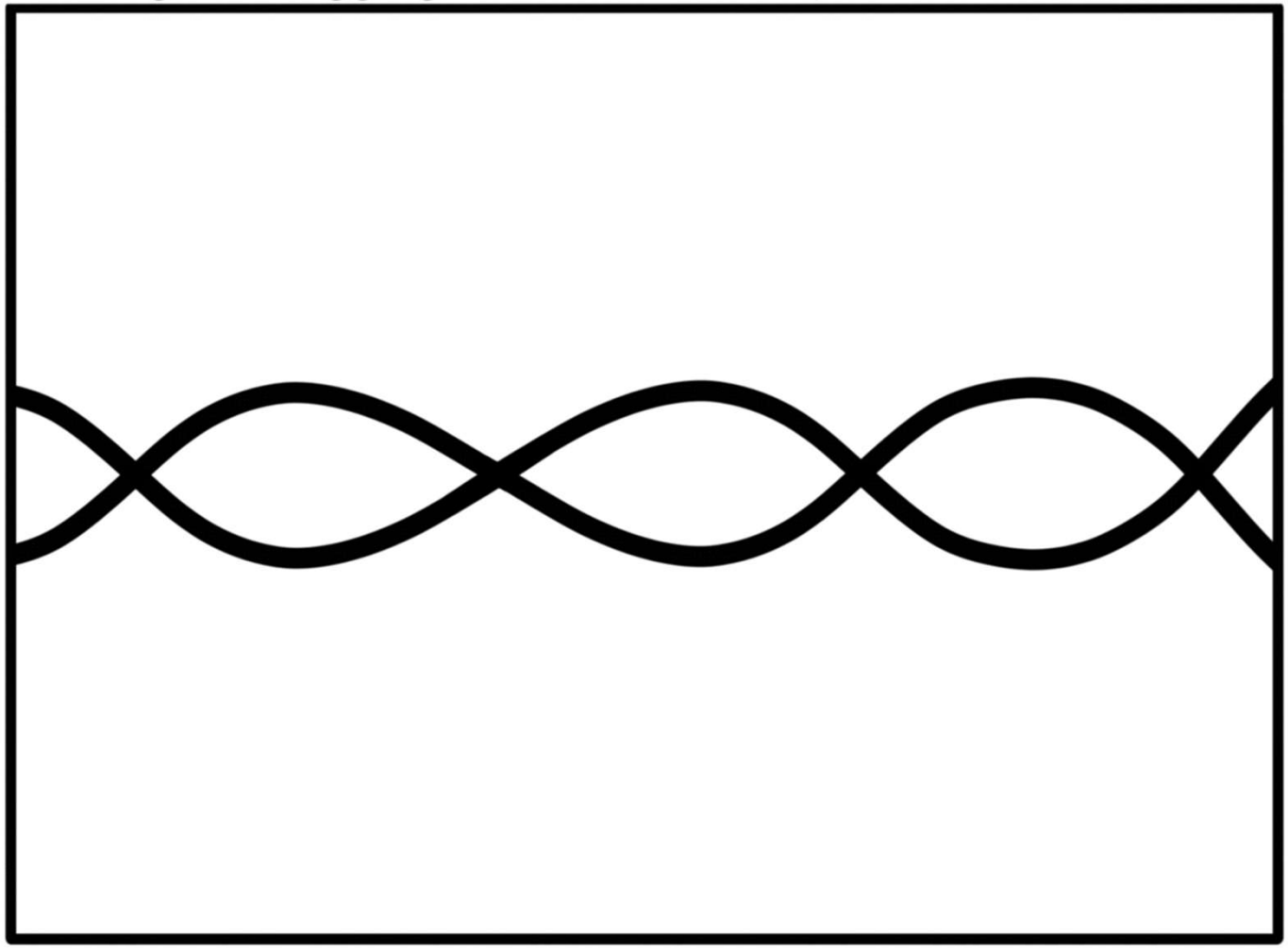
- **termination** in DNA replication, the completion of new DNA strands and dismantling of the replication machine

- As the replication fork progresses along the replicating chromosome, only a very short region of DNA is found in a single-stranded form.
- As soon as the newly formed strands are complete, they rewind automatically into their chemically stable helix structure. Replication proceeds until the new strands are complete and the two new DNA molecules separate from one another.


DNA polymerase
3′
5′
helicase
3′
5′
leading strand
DNA polymerase adding DNA nucleotides to RNA primer
RNA primer
Okazaki fragment
lagging strand
primase
RNA primer removed and replaced with DNA nucleotides
3′
5′
direction of replication fork movement
DNA polymerase
joined by DNA ligase

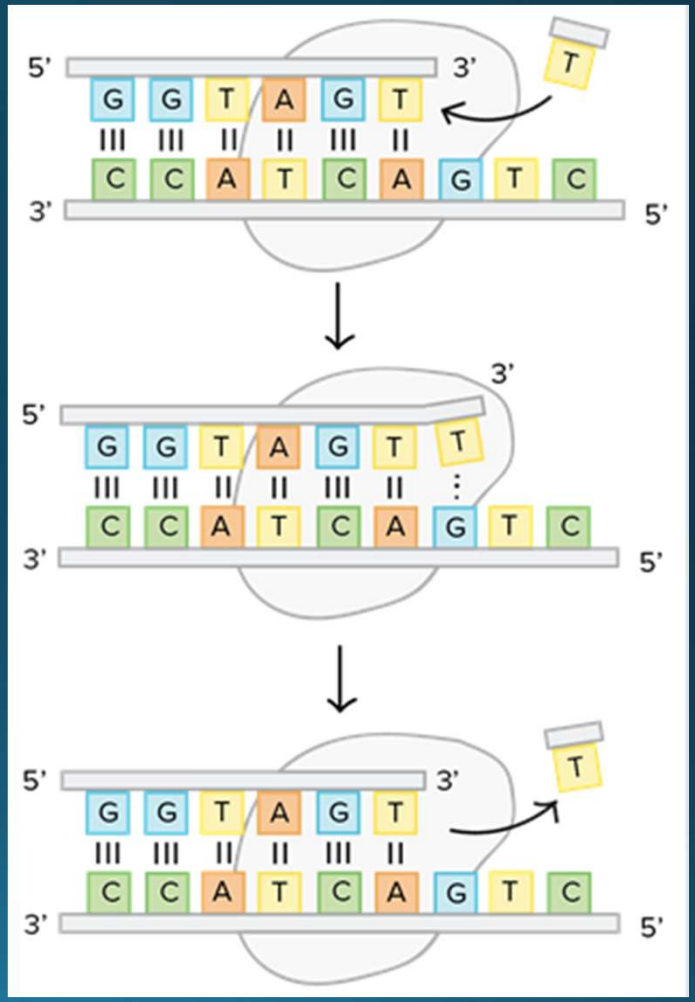
Leading and Lagging Strand in DNA Replication

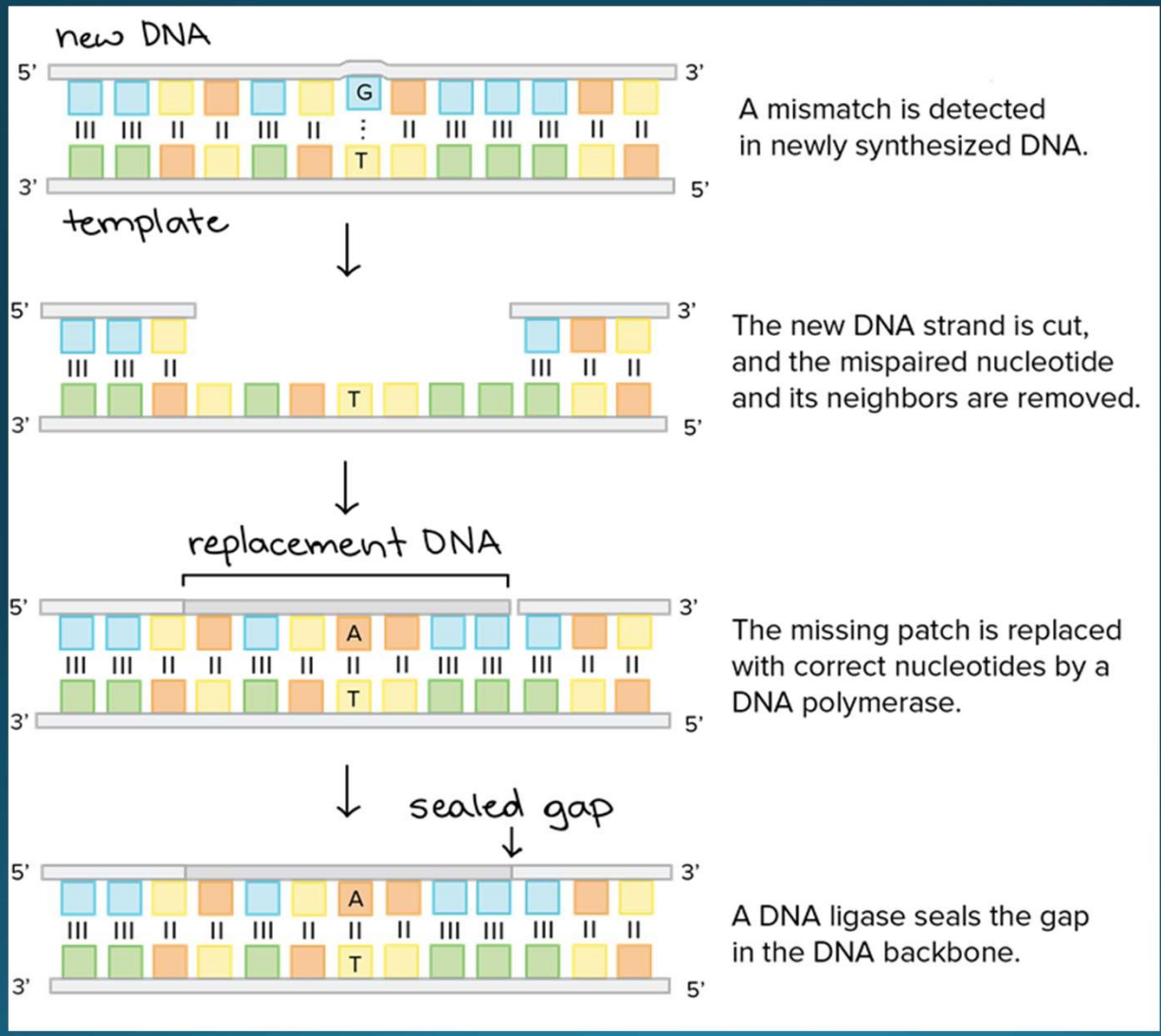
@AmoebaSisters



Proofreading

- **DNA polymerase** has an important proofreading function, as well.
- After each nucleotide is added to a new DNA strand, DNA polymerase can recognize whether or not hydrogen bonding is taking place between the new base and its complement on the original strand.
- **The absence of hydrogen bonding indicates a mismatch between the bases.** When this occurs, DNA polymerase excises the incorrect base from the new strand and **adds the correct base using the parent strand as a template.**





A mismatch is detected in newly synthesized DNA.

The new DNA strand is cut, and the mispaired nucleotide and its neighbors are removed.

The missing patch is replaced with correct nucleotides by a DNA polymerase.

A DNA ligase seals the gap in the DNA backbone.

Example 1

- **Given the Parent DNA sequence ATG – GTA – CGT what is the complementary DNA sequence?**

Remember our pairings from earlier
A-T
C-G

TAC – CAT - GCA

Example 2

- **Given the complementary DNA sequence GCA – AAA – CAC what is the parent DNA sequence?**

Remember our pairings from earlier
A-T
C-G

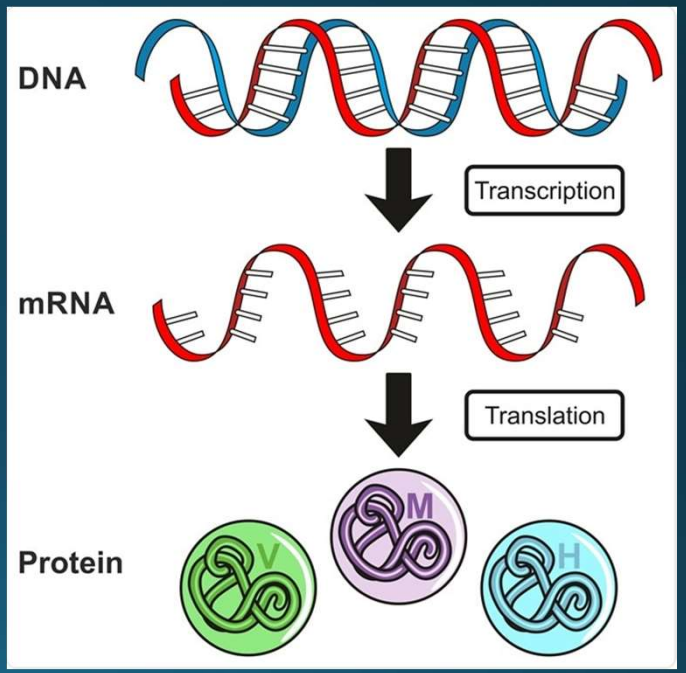
CGT – TTT - GTG

- Students are expected to engage in an engineering design activity to construct a working model of a short DNA strand (8-10 base pairs). The model should show the molecular structure of DNA and be able to simulate the process of DNA replication (Investigation 15.A, *NL Biology*, p. 592).

- Exit Card #9

Gene Expression & The Genetic Code

- **Proteins consist of a sequence of molecules called amino acids.**
- The specific sequence of amino acids determines the chemical properties of each protein.
- A given set of amino acids, arranged in a particular order, could produce the proteins that are responsible for inherited traits
- The genetic code determines how the amino acids are strung together and how the proteins are made.
- The order of the nucleotides in a gene provides the information, written in genetic code, that is necessary to build a protein.



- The genetic code has three important characteristics.

- **1.** The genetic code is *redundant*—that is, more than one codon can code for the same amino acid.
- **2.** The genetic code is *continuous*. That is, the genetic code reads as a series of three-letter codons without spaces, punctuation, or overlap. Knowing exactly where to start and stop translation is essential.
- **3.** The genetic code is nearly *universal*. Almost all living organisms build proteins with the same genetic code.

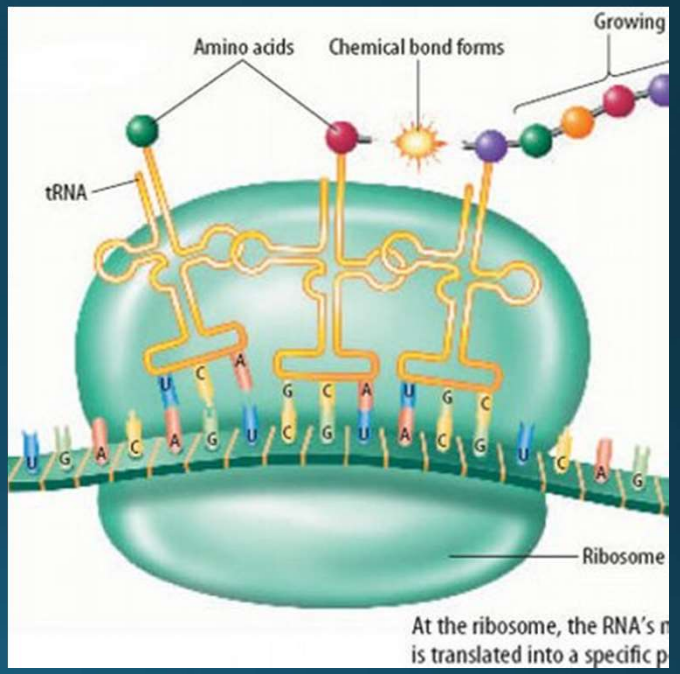
First Position	Second Position: U	Second Position: C	Second Position: A	Second Position: G	Third Position
U	Phenylalanine	Serine	Tyrosine	Cysteine	U
	Phenylalanine	Serine	Tyrosine	Cysteine	C
	Leucine	Serine	Stop	Stop	A
	Leucine	Serine	Stop	Tryptophan	G
C	Leucine	Proline	Histidine	Arginine	U
	Leucine	Proline	Histidine	Arginine	C
	Leucine	Proline	Glutamine	Arginine	A
	Leucine	Proline	Glutamine	Arginine	G
A	Isoleucine	Threonine	Asparagine	Serine	U
	Isoleucine	Threonine	Asparagine	Serine	C
	Isoleucine	Threonine	Lysine	Arginine	A
	Methionine	Threonine	Lysine	Arginine	G
G	Valine	Alanine	Aspartic acid	Glycine	U
	Valine	Alanine	Aspartic acid	Glycine	C
	Valine	Alanine	Glutamic acid	Glycine	A
	Valine	Alanine	Glutamic acid	Glycine	G

Protein Synthesis

- The theory that genetic information flows from DNA to RNA to protein is often referred to as the "**central dogma**" of gene expression.
- Protein Synthesis takes place in two steps.

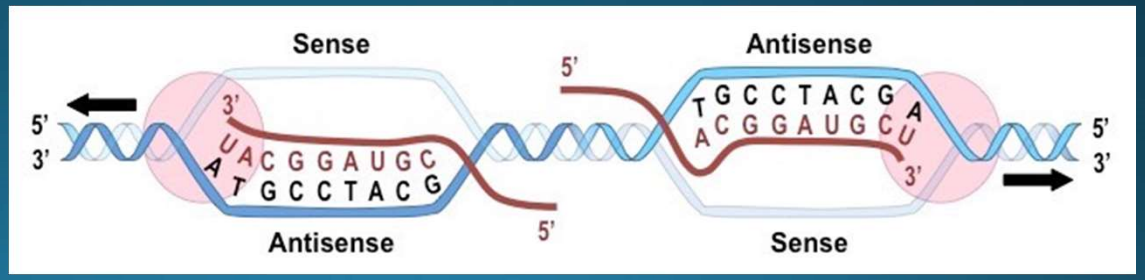
- **1.) Transcription** process of producing RNA from DNA

- **2.) Translation** process of producing a polypeptide based on an mRNA sequence

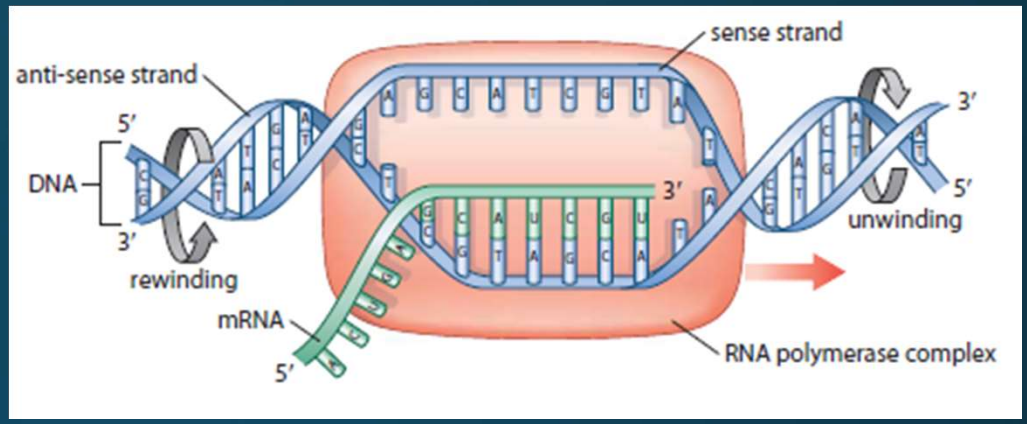


Transcription

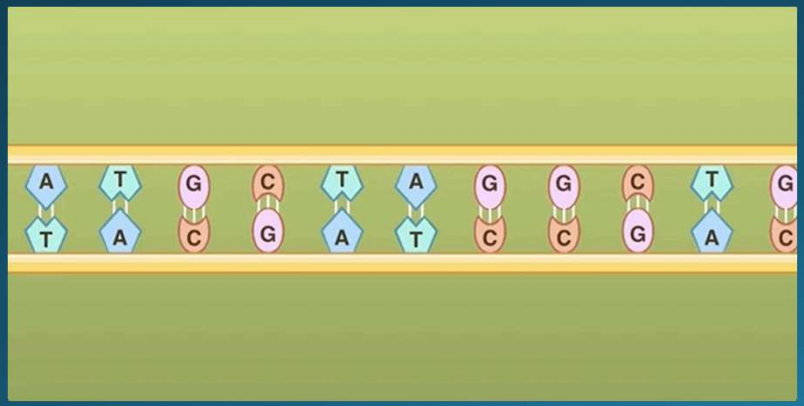
- During transcription, the information in a segment of DNA is copied into messenger RNA (mRNA).
- **messenger RNA (mRNA)** RNA that carries the genetic code from DNA to protein synthesis machinery
- Only one strand of the double-stranded DNA molecule is transcribed.
- This strand is called the ***anti-sense***, or *coding, strand*. The other strand, which is not transcribed, is called the ***sense***, or *non-coding, strand*.

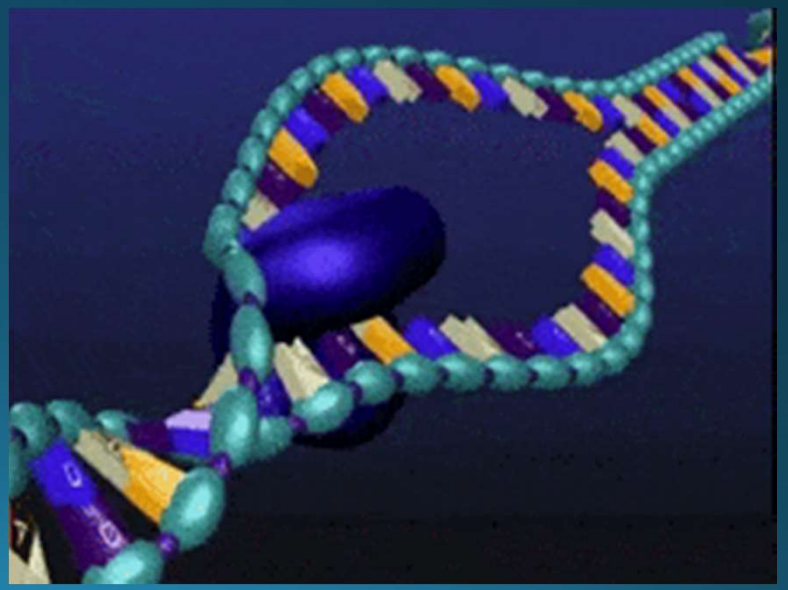


- **RNA polymerase** main enzyme involved in formation of RNA from DNA



- **Codon** set of three bases that code for an amino acid

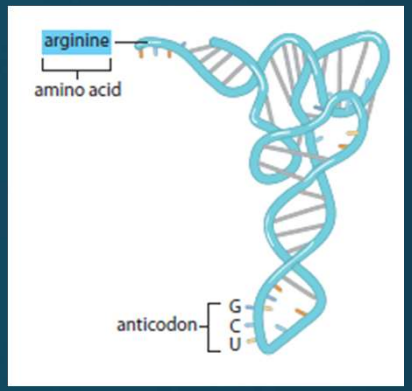


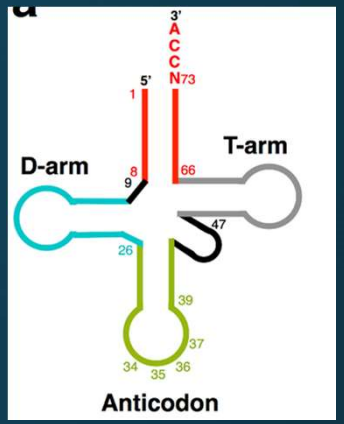


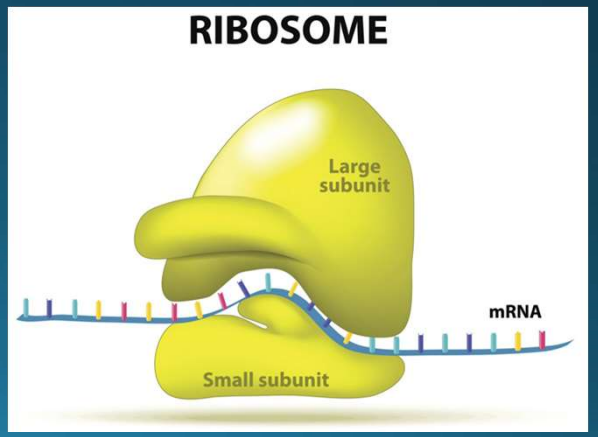
Translation

- For a cell to create the proteins it needs, it must **translate the codons along a stretch of mRNA into amino acid sequences.**
- This process requires both a chemical translator and a set of cellular protein synthesis equipment.
- Once the mRNA reaches the cytoplasm, the translator and protein synthesis equipment work together to assemble the proteins.

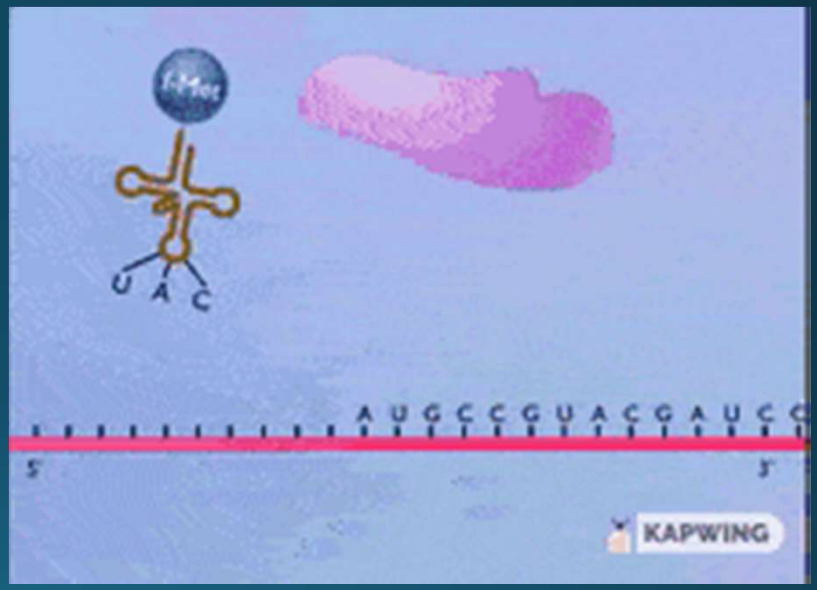
t-RNA





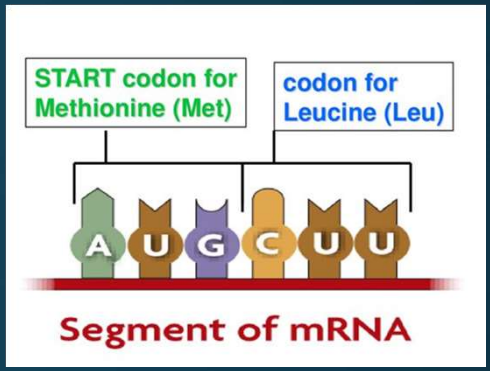


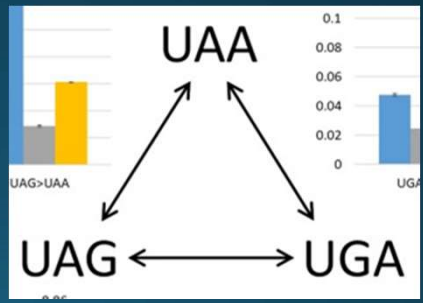
- **transfer RNA (tRNA)** works with mRNA in translation by delivering correct amino acid
- **anticodon** base triplet on tRNA complementary to mRNA codon
- **ribosomal RNA (rRNA)** RNA associated with ribosomes

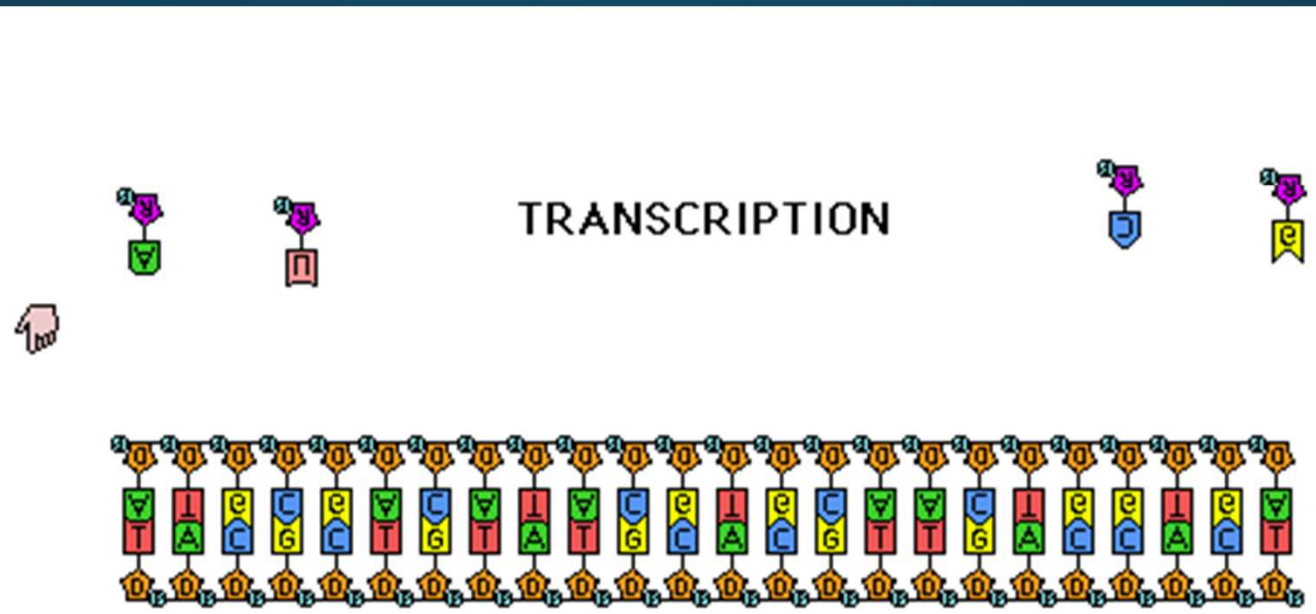


Translation follows a cycle of four steps:

- 1.) The first tRNA molecule, carrying the amino acid **methionine**, base-pairs with the first exposed mRNA codon—the **start codon**,
- 2. A second loaded tRNA molecule arrives at the codon adjacent to the first tRNA.
- 3. Enzymes catalyze the formation of a chemical bond that joins the amino acid carried by the first tRNA to the amino acid carried by the second tRNA. At the same time, the **amino acid chain is transferred from the first tRNA to the second tRNA**.
- 4. The ribosome moves a distance of one codon along the mRNA strand. The first tRNA molecule detaches from the mRNA and **picks up another amino acid**. The second tRNA now holds a growing amino acid chain. A third tRNA molecule arrives at the newly exposed codon next to the second tRNA, and the cycle repeats. The translation cycle continues until a **stop codon** is reached.







(in nucleus)

Types of Codon Tables

- **CODON TABLES USE mRNA ONLY!**

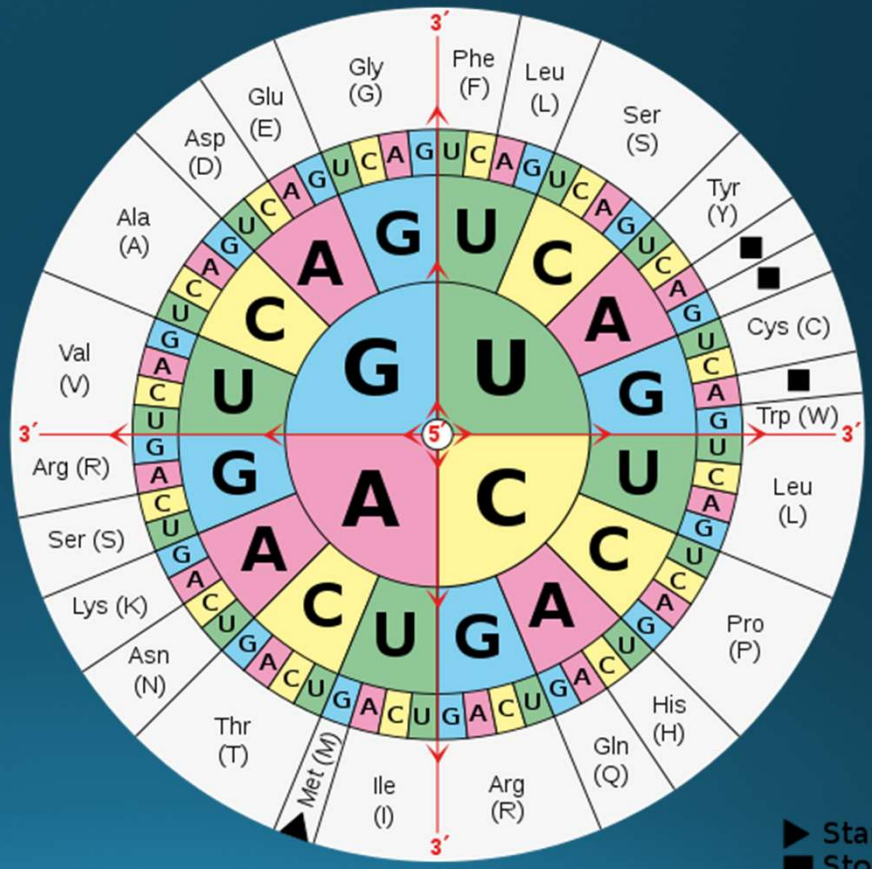
RNA codon table

1st position	2nd position U	2nd position C	2nd position A	2nd position G	3rd position
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

Amino Acids

Ala: Alanine
Arg: Arginine
Asn: Asparagine
Asp:Aspartic acid
Cys:Cysteine
Gln: Glutamine
Glu: Glutamic acid
Gly: Glycine
His: Histidine
Ile: Isoleucine
Leu: Leucine
Lys: Lysine
Met: Methionine
Phe: Phenylalanine
Pro: Proline
Ser: Serine
Thr: Threonine
Trp: Tryptophane
Tyr: Tyrosisne
Val: Valine

- **Start from the middle and work your way out**



- Do not forget that mRNA and tRNA do not have thymine

- DNA has A T G C
- A – T
- G – C

- RNA has A U G C

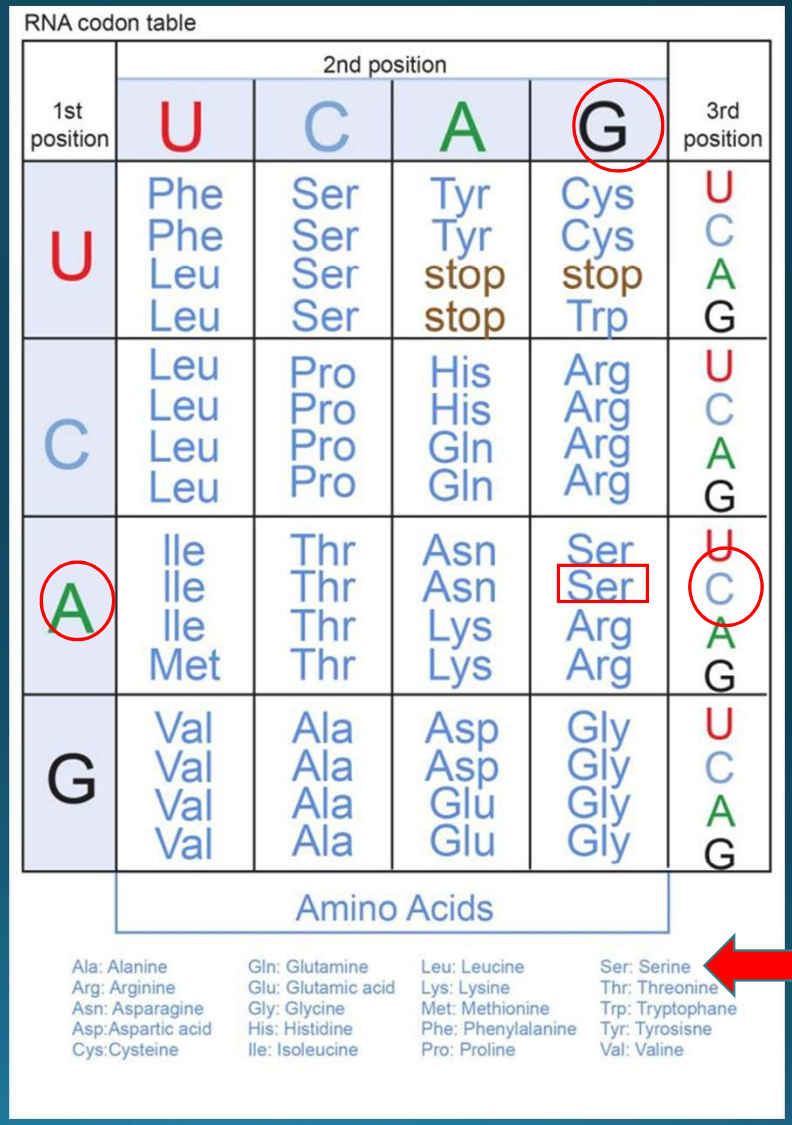
- A – U
- G – C

Example 1

- What amino acid does the **CODON** sequence AGC code?

Codon means that it is mRNA already transcribed, so we just go to our table and find the translation.

Serine



Example 2

- What amino acid does the **DNA** sequence ATG code?

Since this is DNA we have to transcribe our mRNA first before we translate it.

Original DNA: ATG

Complementary mRNA Sequence: UAC

Now we use the mRNA table

Tyrosisne

RNA codon table

1st position	2nd position U	2nd position C	2nd position A	2nd position G	3rd position
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

Amino Acids

Ala: Alanine
Arg: Arginine
Asn: Asparagine
Asp:Aspartic acid
Cys:Cysteine
Gln: Glutamine
Glu: Glutamic acid
Gly: Glycine
His: Histidine
Ile: Isoleucine
Leu: Leucine
Lys: Lysine
Met: Methionine
Phe: Phenylalanine
Pro: Proline
Ser: Serine
Thr: Threonine
Trp: Tryptophane
Tyr: Tyrosisne
Val: Valine

Example 3

- What amino acid does the **ANTICODON** sequence AUU code?

Since this is tRNA we have to figure out our mRNA first before we translate it.

Original tRNA: AUU

Complementary mRNA Sequence: UAA

Now we use the mRNA table

STOP

RNA codon table

1st position	2nd position U	2nd position C	2nd position A	2nd position G	3rd position
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

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Gln: Glutamine
Glu: Glutamic acid
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Leu: Leucine
Lys: Lysine
Met: Methionine
Phe: Phenylalanine
Pro: Proline
Ser: Serine
Thr: Threonine
Trp: Tryptophane
Tyr: Tyrosisne
Val: Valine

Example 4

- If the polypeptide sequence, phenylalanine - isoleucine - threonine, were produced through transcription, what mRNA sequence was present originally
- (A) AAA UAA UGG
- (B) AAG UAU AAU
- (C) UUC AUG ACA
- (D) UUU AUU ACC

mRNA for phenylalanine are
UUU
UUC

This means that B or D could be right, so we now have to check isoleucine

To do this question we have to first look at the mRNA sequences that created those amino acids and make a list

mRNA for isoleucine are
AUU
AUC
AUA

D has to be right

RNA codon table

1st position	2nd position U	2nd position C	2nd position A	2nd position G	3rd position
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

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Glu: Glutamic acid
Gly: Glycine
His: Histidine
Ile: Isoleucine
Leu: Leucine
Lys: Lysine
Met: Methionine
Phe: Phenylalanine
Pro: Proline
Ser: Serine
Thr: Threonine
Trp: Tryptophane
Tyr: Tyrosisne
Val: Valine

Example 5

- Using the codon table, which **DNA** sequence was used as a template to produce the polypeptide sequence glycine - isoleucine - phenylalanine?
- (A) CCC TAG AAC
- (B) CCG TAA AAG
- (C) GGA TAC AAT
- (D) GGC TAT AAA

To do this question we have to first look at the mRNA sequences that created those amino acids and convert them to DNA

mRNA for glycine are
GGU
GGC
GGA
GGG

DNA for glycine are
CCA
CCG
CCT
CCC

This means that A or B could be right, so we now have to check isoleucine

mRNA and DNA for isoleucine are
AUU = TAA
AUC = TAG
AUA = TAT

A has to be right

RNA codon table

1st position	2nd position U	2nd position C	2nd position A	2nd position G	3rd position
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

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- Complete Activities 15.2 and 15.3 (*NL Biology*, pp. 597-98).

- INVESTIGATION
- Simulating Protein Synthesis p 600

- Exit Card #10