UNIT II

- Biological Diversity
- Chemistry of life: chemical bonds
- Biochemistry and Human biology
- Protein synthesis
- Stem cells and tissue engineering

BIODIVERSITY

Biological diversity - or biodiversity - is the term given to the varieties of life on Earth. It is the result of billions of years of evolution, shaped by natural processes and, increasingly, by the influence of humans.

It forms the web of life of which we are an integral part and upon which we so fully depend.

BIODIVERSITY

1) **Variety of species**
2) **Genetic differences**
3) **Variety of ecosystems**

Genetic diversity

Taxonomic diversity

Community or ecosystem diversity

WHY IS BIODIVERSITY IMPORTANT FOR YOU AND THE WORLD?

Protecting biodiversity is in our self interest, providing the goods and services that sustain our lives including:

- Provision of shelter and building materials
- Stabilization and moderation of the Earth's climate
- Purification of air and water
- Provision of food, fuel, and fibre
- Cultural and aesthetic benefits, etc.

WHAT ARE THE CHALLENGES FACING BIODIVERSITY?

Species have been disappearing at up to 1000 times the natural rate

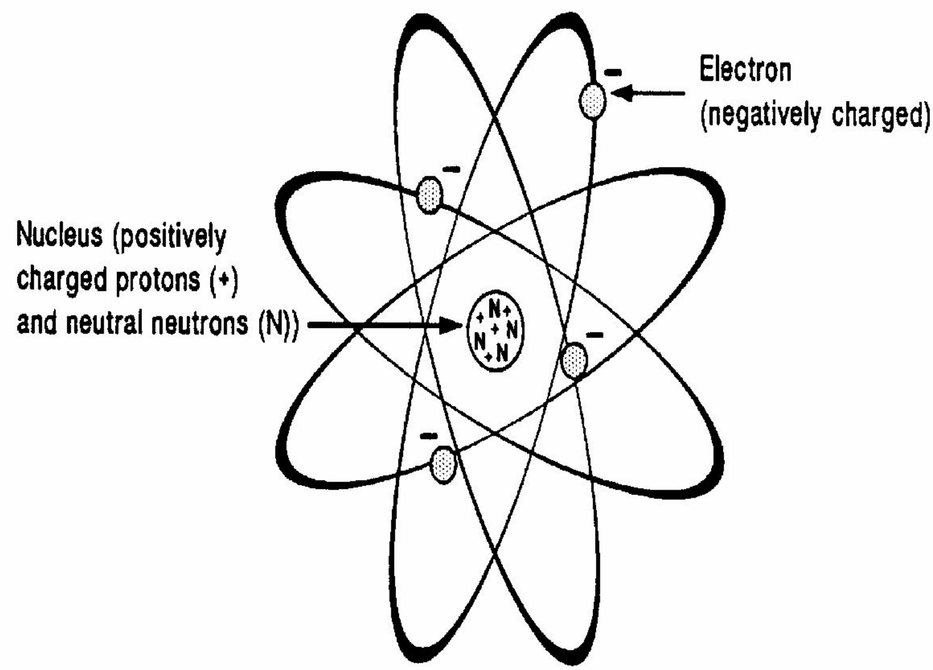
- **An estimated 34,000 plant and 5,200 animal species face extinction, including one in eight birds and one third of all amphibians**
- **20% of known bird species have already disappeared**
- **41% of mammals are in decline and 28% are under direct threat**
- **45% of the Earth's original forests are gone. Forest areas of about four times the size of Belgium are being lost every year.**

Human activities are creating the greatest wave of extinction since the natural disaster that wiped out the dinosaurs 65 million years ago

Chemistry of life: chemical bonds

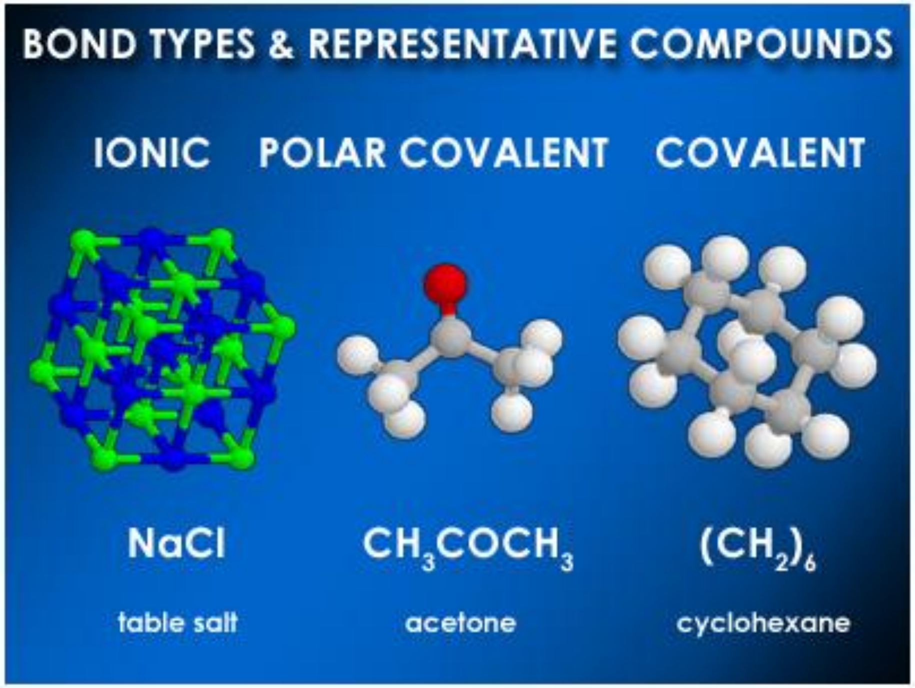
Chemistry of Life

- All matter is built up of simple units called atoms.
- Although the word atom means something that cannot be cut (a = "without," tom = "cut"), these elementary particles are actually made up of many smaller parts, which are themselves further divisible.
- Elements are substances that consist of the same kinds of atoms.
- Compounds consist of units called molecules, which are intimate associations of atoms (in the case of compounds, different atoms) joined in precise arrangements.

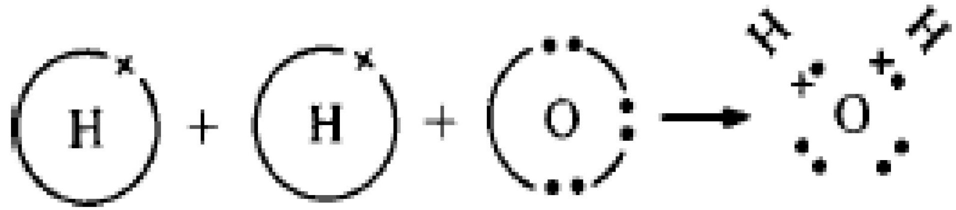


- **Atoms interact with one another to form chemical communities. The tightly knit atoms making up the communal molecules are held together by chemical bonding.**

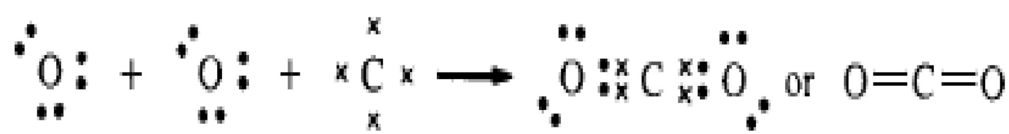
- **One way of achieving this more stable state is for an atom with very few electrons in its outer shell to donate them to an atom with an outer shell that is almost complete.**
  - **The atom that donates the electrons will then have more protons than electrons and assume a positive charge; it is called a cation. The atom receiving the electrons assumes a negative charge and is called an anion.**
  - **These two oppositely charged ions are electrostatically attracted to each other and are said to have an ionic, or polar, bond.**



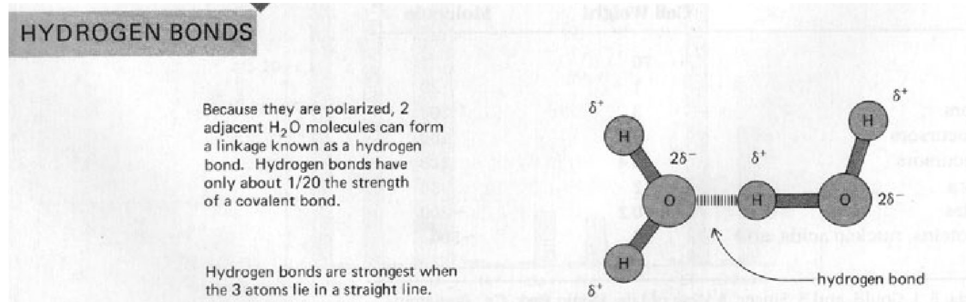
- A second way in which atoms may join with one another to bring about a filling of their outermost shells is by sharing a pair of electrons.
  - The two bonding atoms provide one electron each in creating the shared pair. This pair of electrons forms a covalent bond that holds the two atoms together. It is represented by a solid line in the formula of a compound.

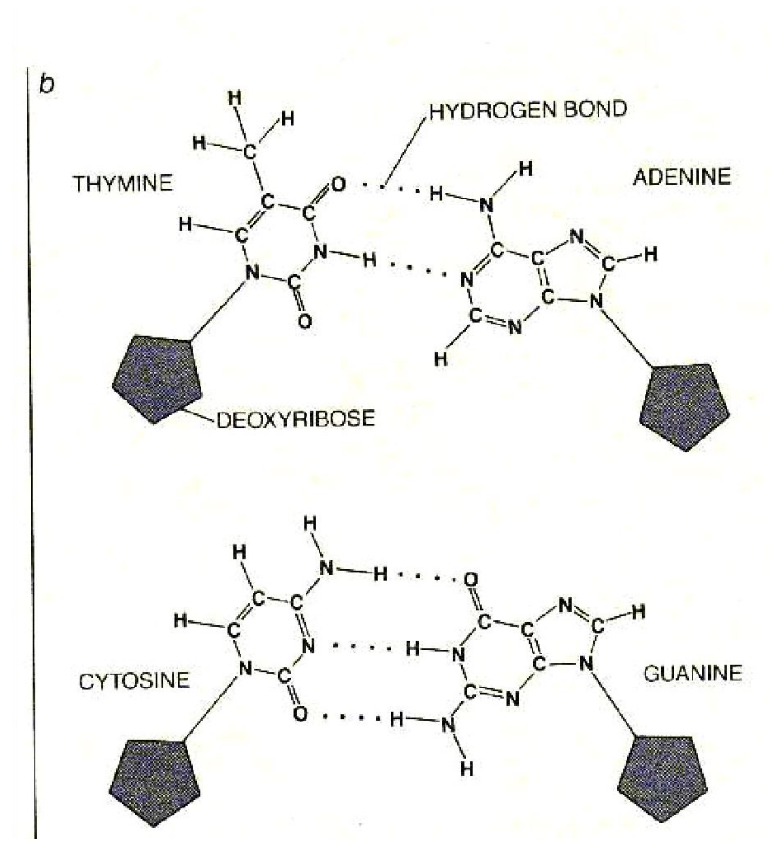


- In many molecules, covalent bonding may occur not just singly (sharing a single pair of electrons), but may involve the formation of double or triple bonds in which two and even three pairs of electrons are shared.
  - These double and triple bonds tend to fix the position of the participating atoms in a rigid manner.



- **Non-covalent bonds (ionic, hydrogen)** are much weaker than **covalent bonds** (electron sharing) and so protein shape can be disrupted especially by temperature, pH , ions (salt).
- It involves more dispersed variations of electromagnetic interactions.
- Critical in maintaining the three-dimensional structure of large molecules, such as proteins and nucleic acids
- There are four commonly mentioned types of non-covalent interactions: hydrogen bonds, ionic bonds, van der Waals forces, and hydrophobic interactions.
  - The noncovalent interactions hold together the two strands DNA in the double helix, stabilize secondary and tertiary structures of proteins, and enable enzyme-substrate binding and antibody-antigen association.





Biochemistry and Human biology

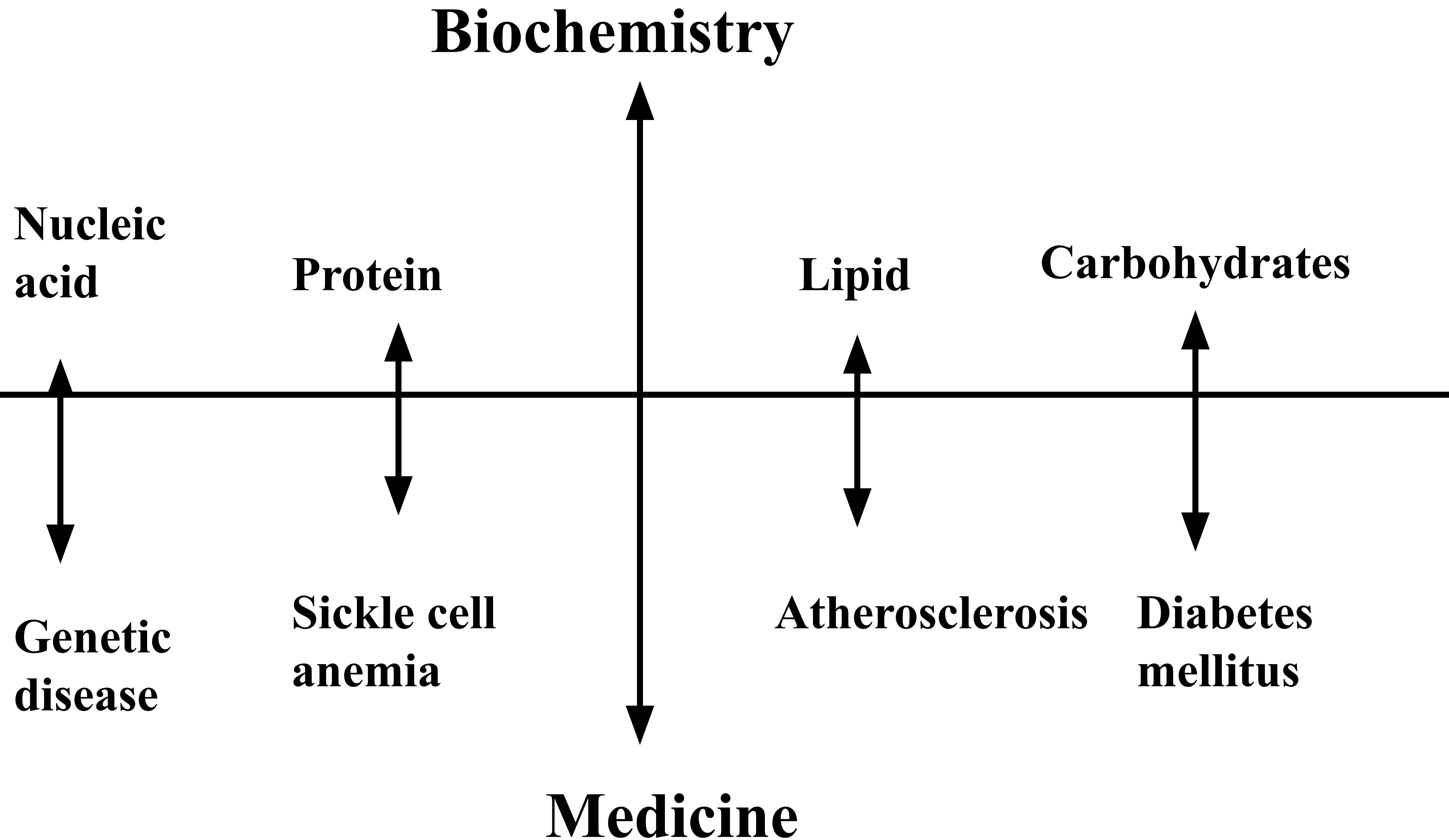
Biochemistry: Where Chemistry & Biology Meet

- Living things require millions of chemical reactions just to survive.
- Metabolism = all the chemical reactions occurring in the body.
- Organic molecules:
  - usually associated with living things.
  - always contain CARBON.
  - are “large” molecules, with many atoms
  - always have covalent bonds (share electrons)

Biochemistry and Human Biology

- **Biochemistry**: Science concerned with the chemical constituents of living cells and with the reaction and process that they undergo.
  - Complete understanding at the molecular level of all the chemical processes associated with living cells
  - An appreciation of the biochemistry of less complex form of life is often direct relevance to human biochemistry

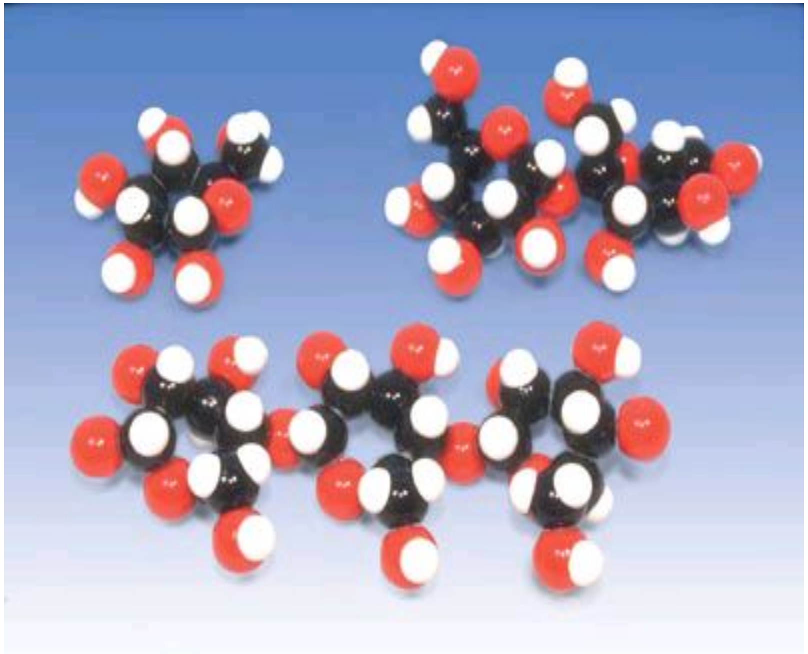
- **Reciprocal relationship between biochemistry and medicine has stimulated mutual advance**
  - Biochemistry studies have illuminated many aspects of health & disease


Biochemistry
Nucleic acid
Protein
Lipid
Carbohydrates
Genetic disease
Sickle cell anemia
Atherosclerosis
Diabetes mellitus
Medicine

S. No.	Disease	Causes
1	Scurvy rickets	deficiencies of vitamins C and D respectively
2	Atherosclerosis	genetic, dietary, environmental factors
3	Cystic fibrosis	mutation in the gene coding the CFTR protein (Cystic fibrosis transmembrane conductance regulator, a protein involved in the transport of chloride ions across cell membranes)
4	Cholera	exotoxin of vibrio cholera
5	Diabetes mellitus type I	genetic and environmental factors resulting in deficiency of insulin
6	Phenylketonuria	mainly mutation in the gene coding phenylalanine hydroxylase

Carbon-based Molecules

- Although a cell is mostly water, the rest of the cell consists mostly of carbon-based molecules

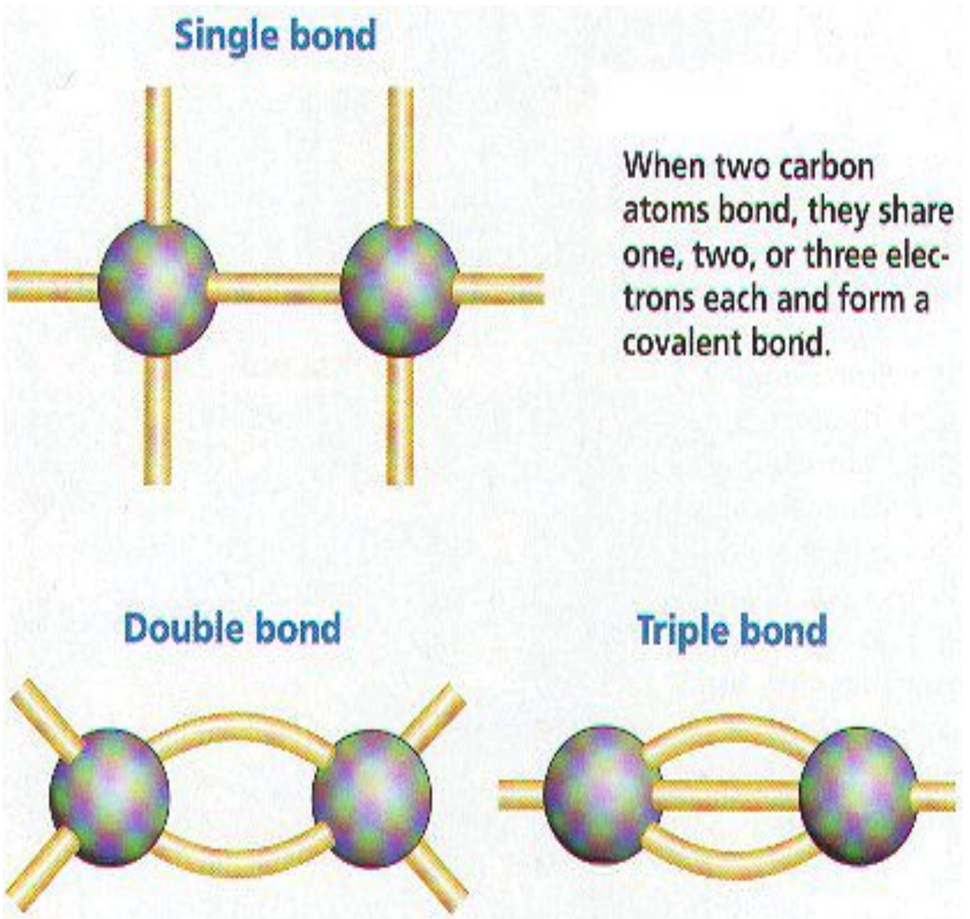


Organic chemistry is the study of carbon compounds

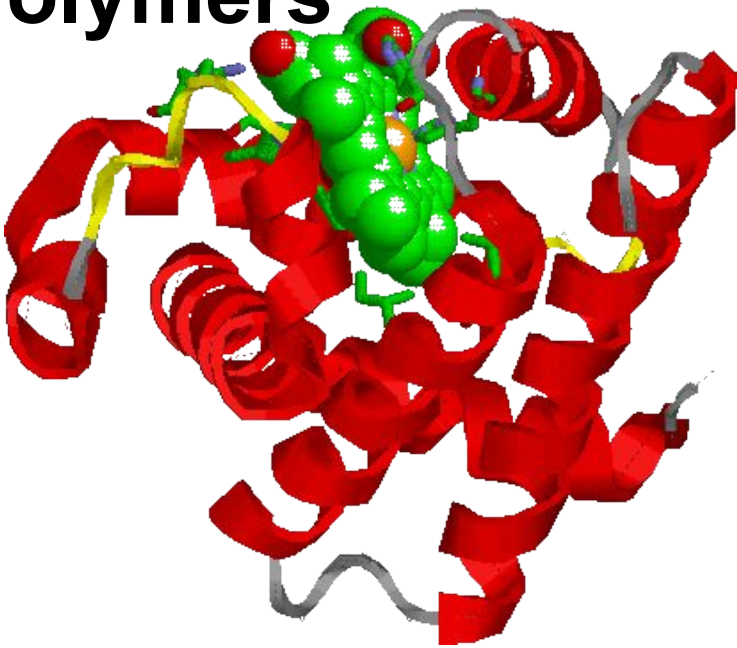
Carbon is a Versatile Atom

- It has four electrons in an outer shell that holds eight

Carbon can share its electrons with other atoms to form up to four covalent bonds



Giant Molecules - Polymers



- Large molecules are called polymers
- Polymers are built from smaller molecules called monomers
- Biologists call them macromolecules

Macromolecules in Organisms

- There are four categories of large molecules in cells:

Carbohydrates

Lipids

Proteins

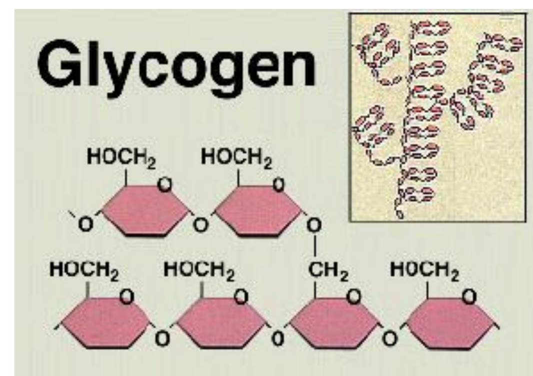
Nucleic Acids

Examples of Polymers

- Proteins

Lipids

Carbohydrates



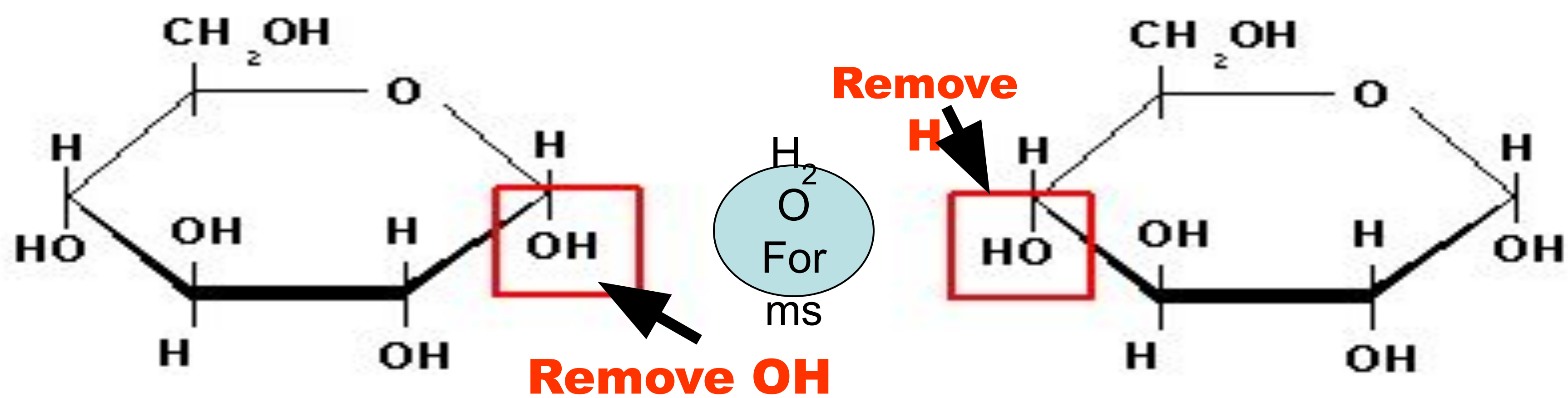
Nucleic Acids

Carbohydrates

- **Carbohydrates include:**
  - –Small sugar molecules in soft drinks
  - –Long starch molecules in rice, wheat, pasta and potatoes

Linking Monomers

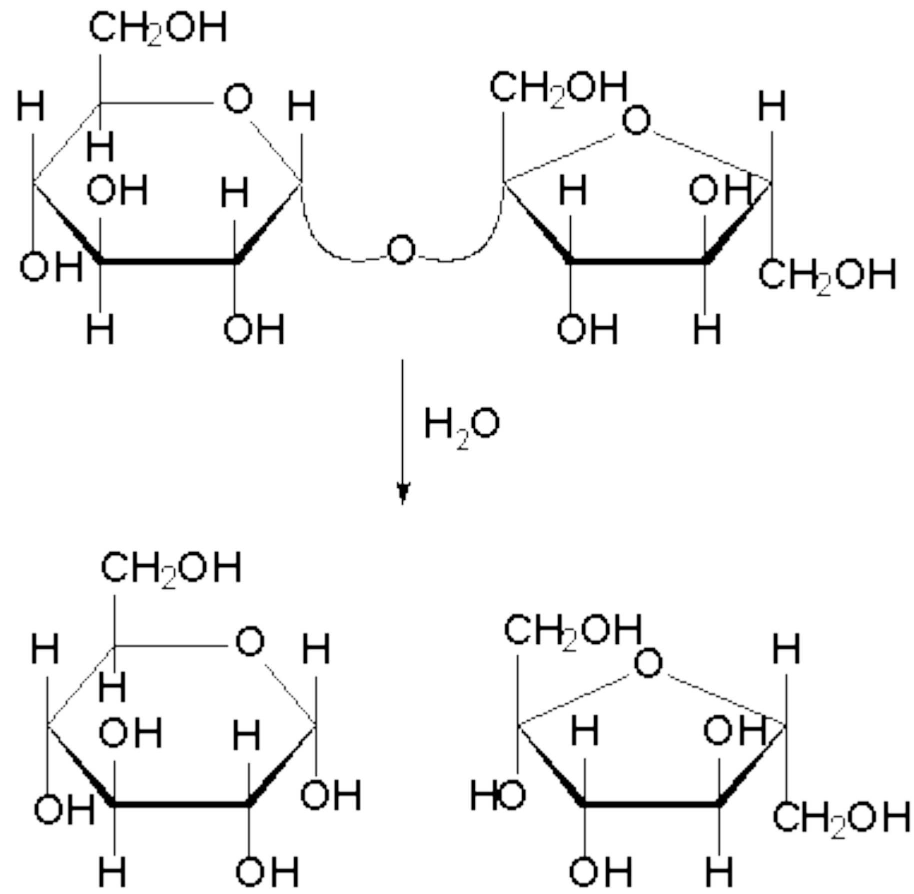
Cells link monomers by a process called condensation or dehydration synthesis (removing a molecule of water)



This process joins two sugar monomers to make a double sugar

Breaking Down Polymers

- **Cells break down macromolecules by a process called hydrolysis (adding a molecule of water)**



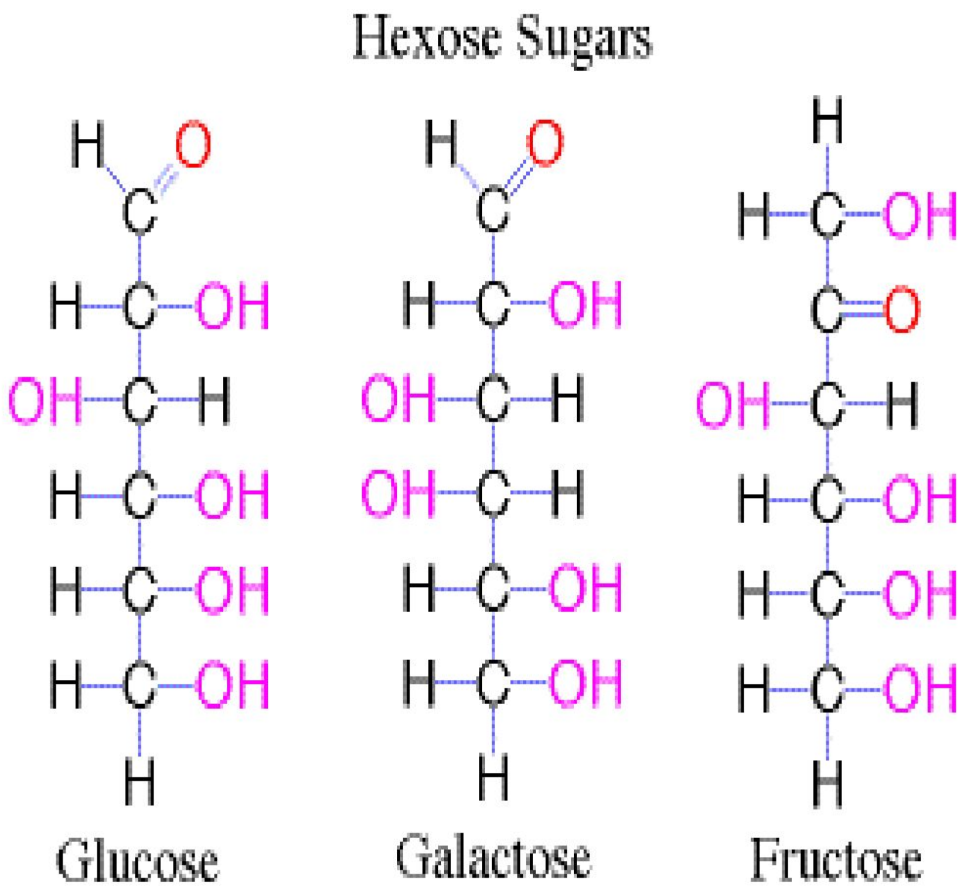
Water added to split a double sugar

Monosaccharides

- Called simple sugars

Include glucose, fructose, & galactose

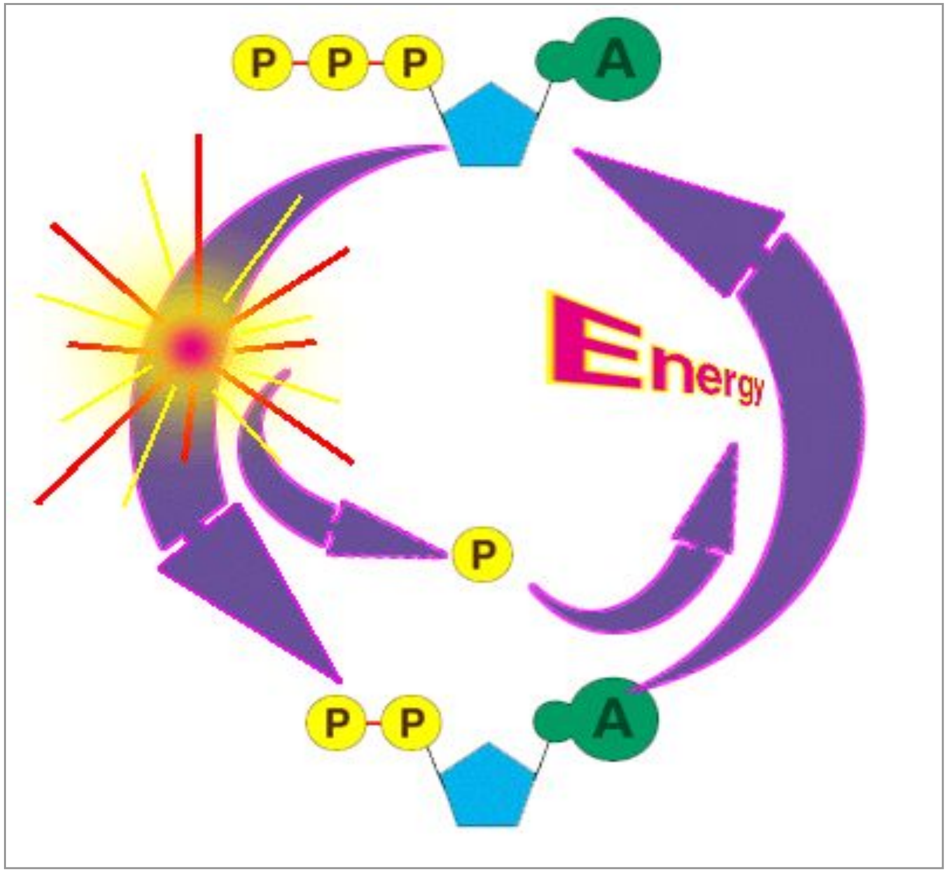
Have the same chemical, but different structural formulas



$C_6H_{12}O_6$

Cellular Fuel

- Monosaccharides are the main fuel that cells use for cellular work



ATP

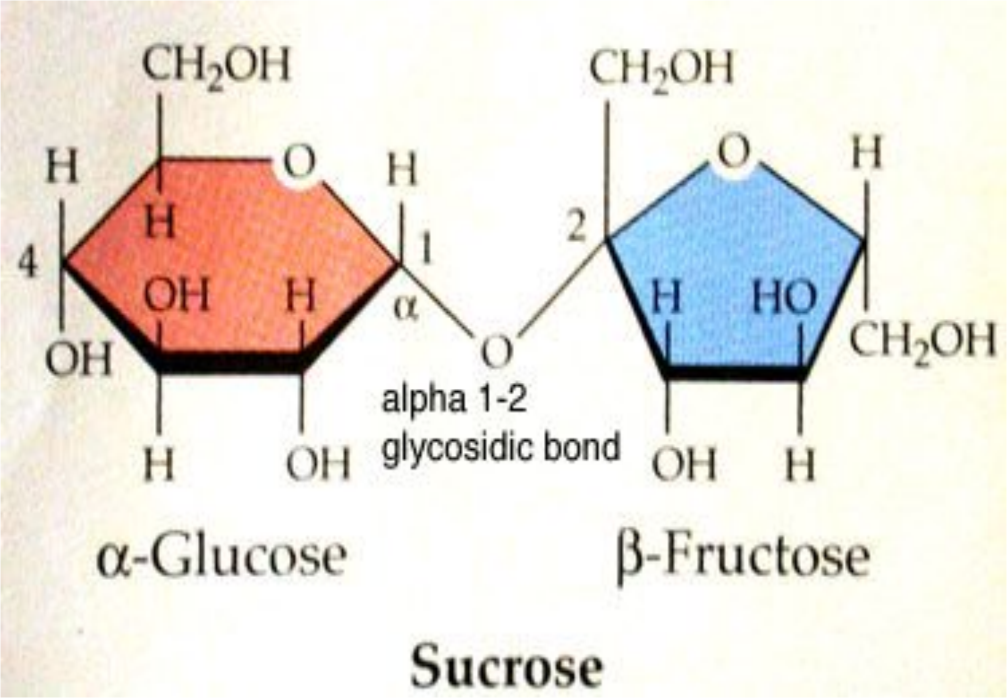
Disaccharides

- A disaccharide is a double sugar.

  They're made by joining two monosaccharides

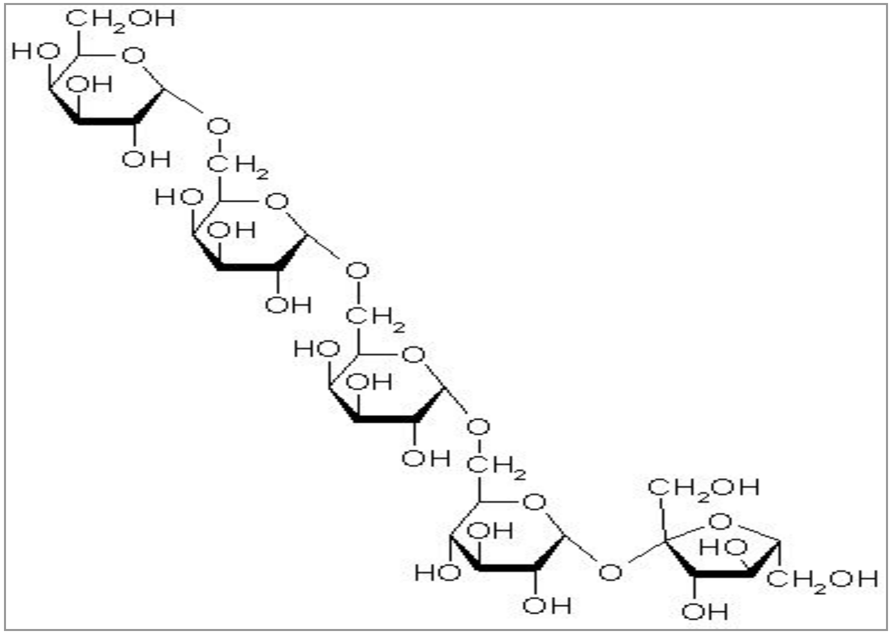
  Involves removing a water molecule (condensation)

  H from sugar and hydroxyl group from other sugar



Bond called a GLYCOSIDIC bond

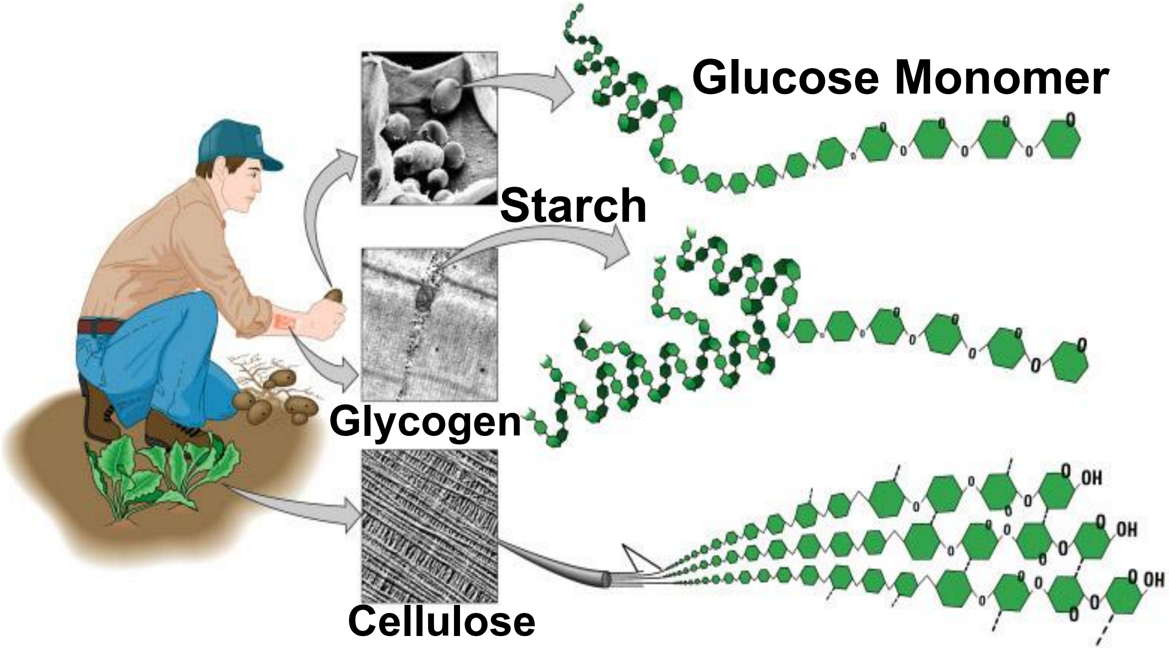
Polysaccharides



- **Complex carbohydrates**

Composed of many sugar monomers linked together

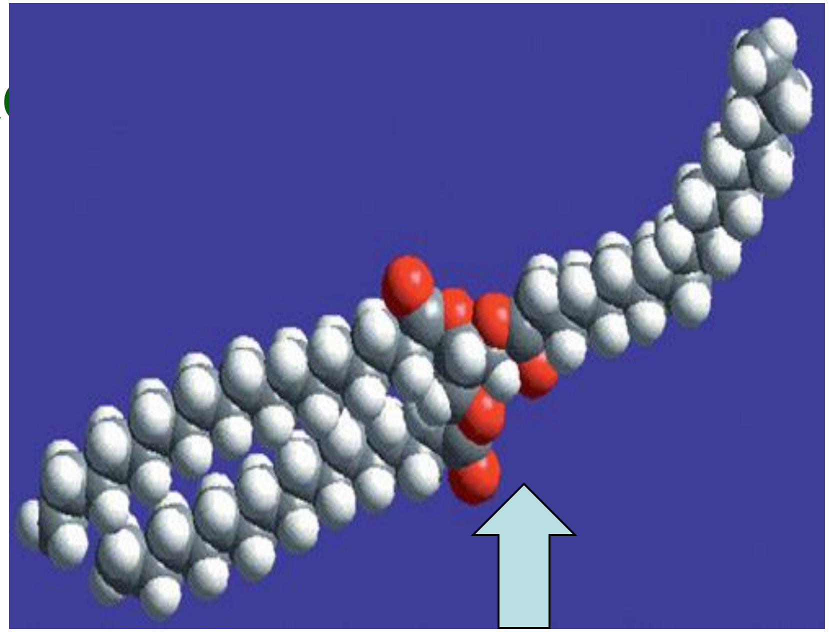
Polymers of monosaccharide chains



Lipids

- Lipids are hydrophobic –"water fearing"
- Hydrophobic tail & little hydrophilic carboxyl head
- Do NOT mix with water
- Includes fats, waxes, steroi

•Fats **store energy**, help to **insulate the body**, and cushion and protect organs



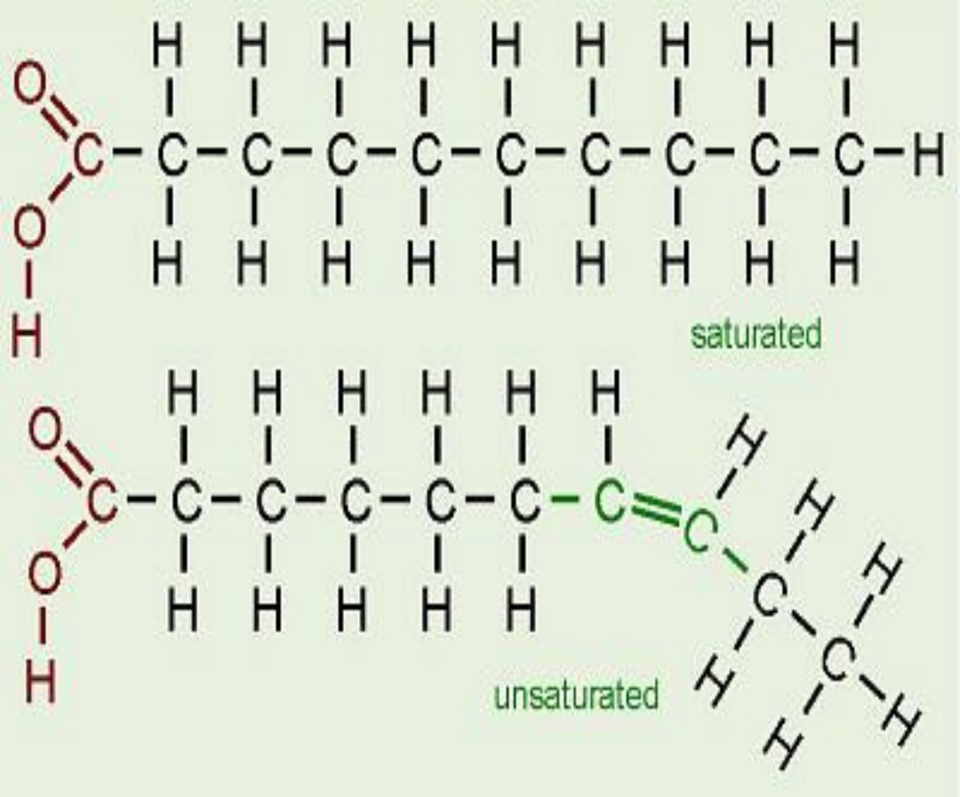
FAT MOLECULE

Types of Fatty Acids

Saturated fatty acids have the maximum number of hydrogens bonded to the carbons (all single bonds between carbons)

Unsaturated fatty acids have less than the maximum number of hydrogens bonded to the carbons (a double bond between carbons)

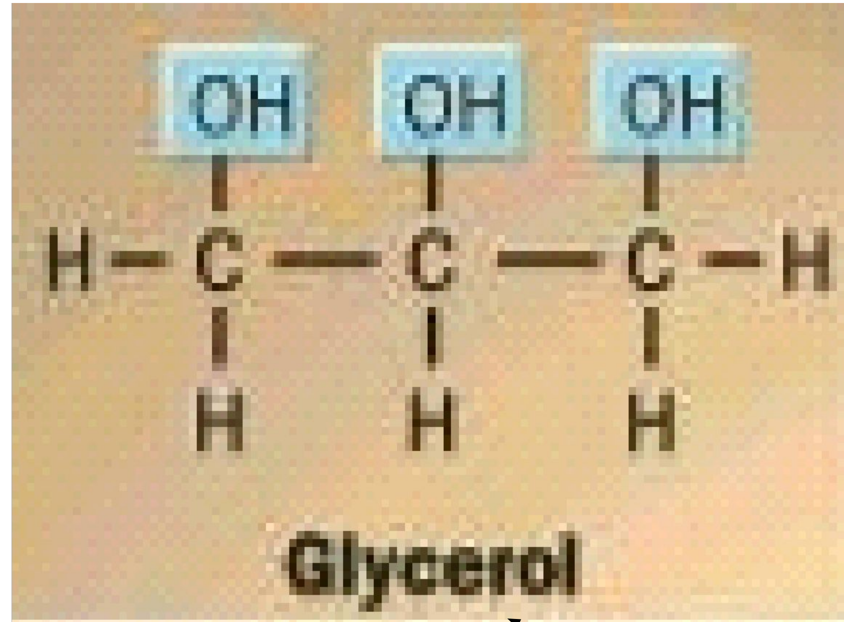
Single Bonds in Carbon chain



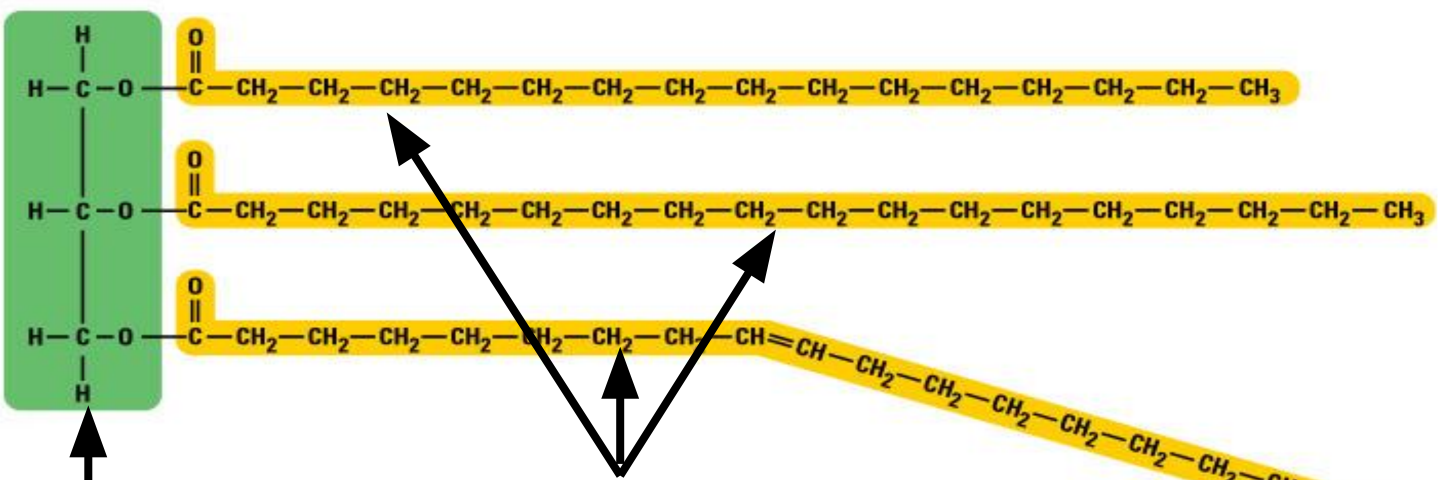
Double bond in carbon chain

Triglyceride

- Monomer of lipids
- **Composed of Glycerol & 3 fatty acid chains**
- **Glycerol forms the “backbone” of the fat**



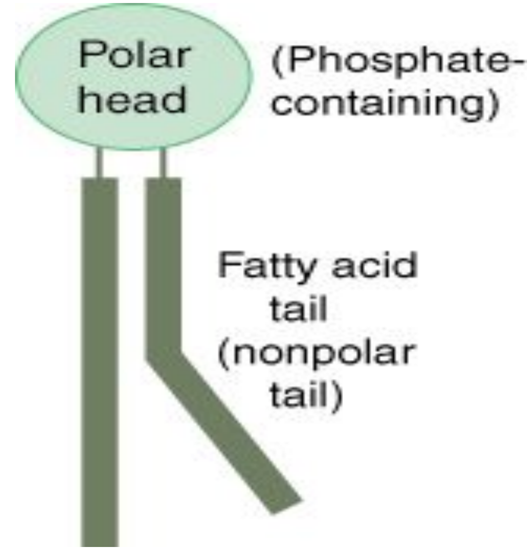
Organic Alcohol
(-OL ending)

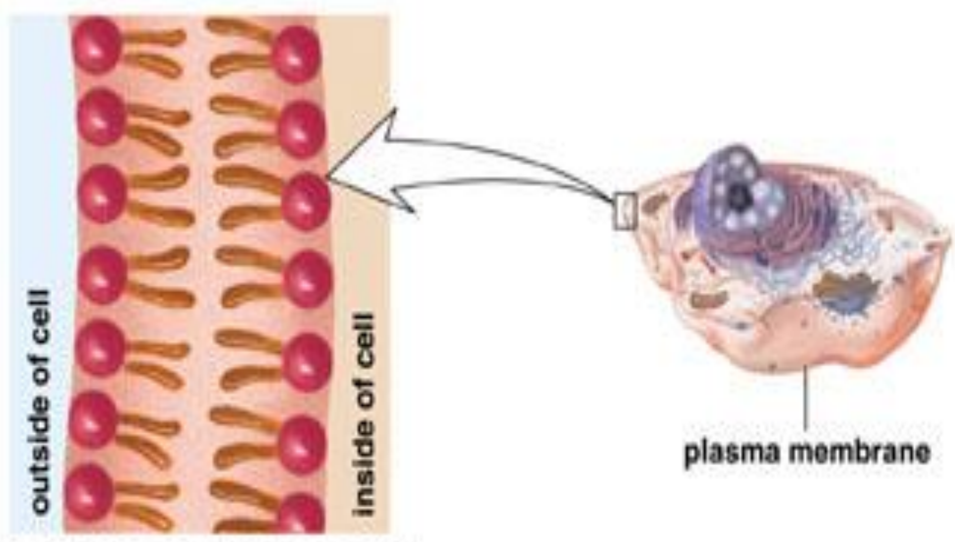


Glycerol **Fatty Acid Chains**

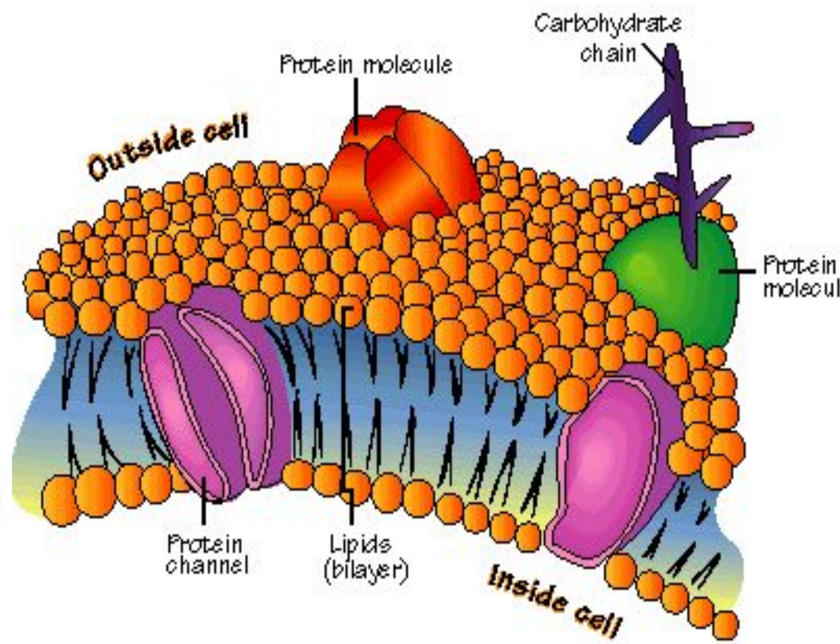
Lipids & Cell Membranes

- Cell membranes are made of lipids called phospholipids
- Phospholipids have a head that is polar & attract water (hydrophilic)
- Phospholipids also have 2 tails that are nonpolar and do not attract water (hydrophobic)





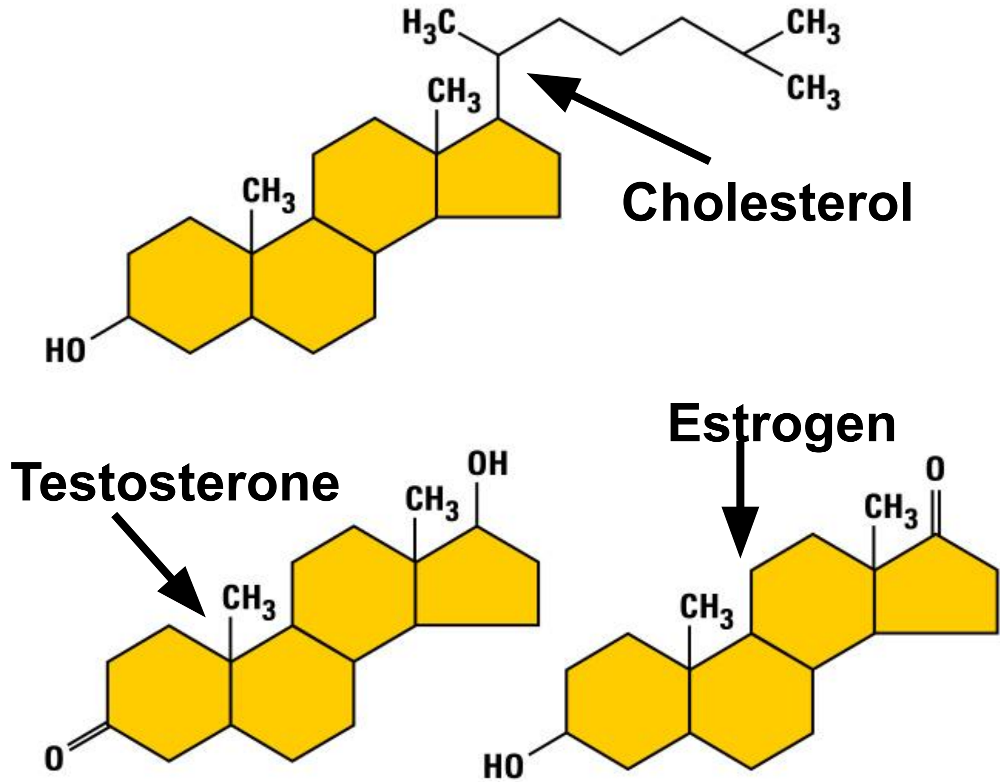
b. Plasma membrane of a cell



Cell membrane with proteins & phospholipids

Steroids

- The carbon skeleton of steroids is bent to form 4 fused rings
- Cholesterol is the “base steroid” from which your body produces other steroids
- Estrogen & testosterone are also steroids



Synthetic Anabolic Steroids

- They are variants of testosterone
- Some athletes use them to build up their muscles quickly
- They can pose serious health risks

Waxes

- A wax is a lipid because of its nonpolar solubility characteristics as well as its extremely hydrophobic (water-hating) properties.
- Waxes are composed of a single, highly complex alcohol joined to a longchain fatty acid in a typical ester linkage.
- Waxes are important structural lipids often found as protective coatings on the surfaces of leaves, stems, hair, skin, etc.
- They provide effective barriers against water loss and in some situations make up the rigid architecture of complex structures such as the honeycomb of the beehive.
- They serve a commercial use as well, in furniture polish, automobile coating compounds, and floor finishes.

Proteins

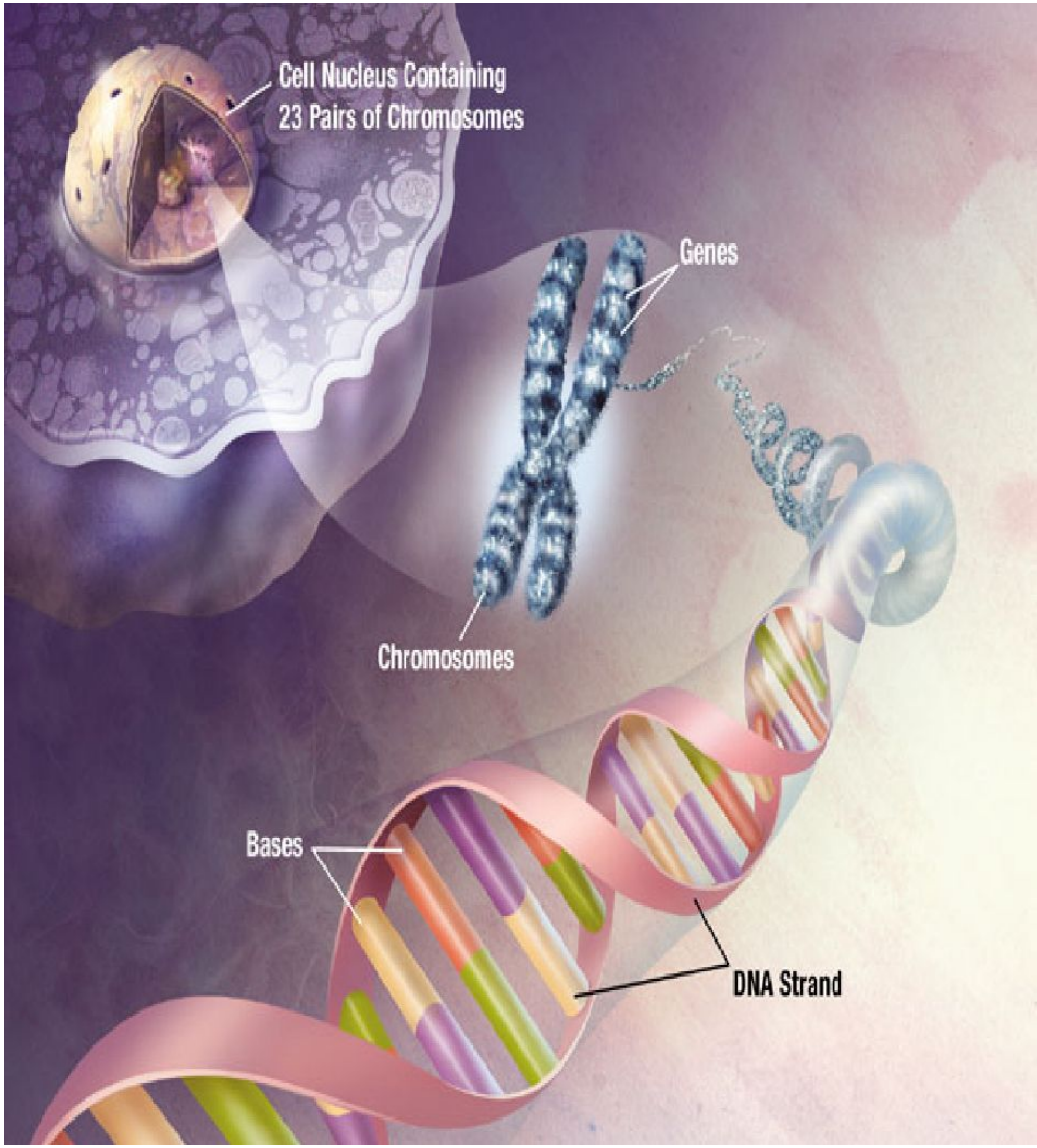
- **Proteins are polymers made of monomers called amino acids**

All proteins are made of 20 different amino acids linked in different orders

Proteins are used to build cells, act as hormones & enzymes, and do much of the work in a cell

Nucleic Acids

- Store hereditary information
  - **Contain information for making all the body's proteins**
  - **Two types exist --- DNA & RNA**

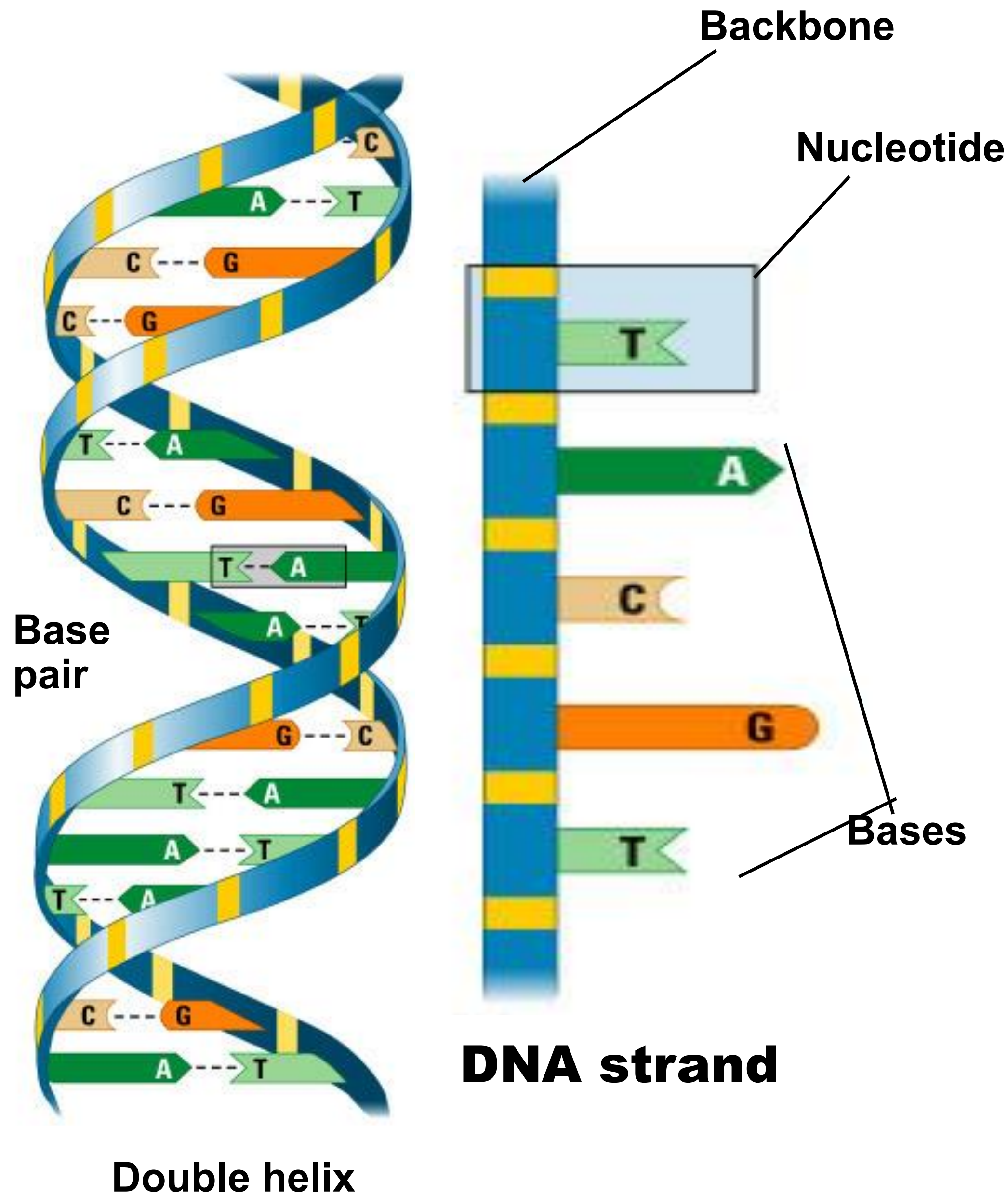


DNA-Deoxyribonucleic acid

- Two strands of DNA join together to form a double helix

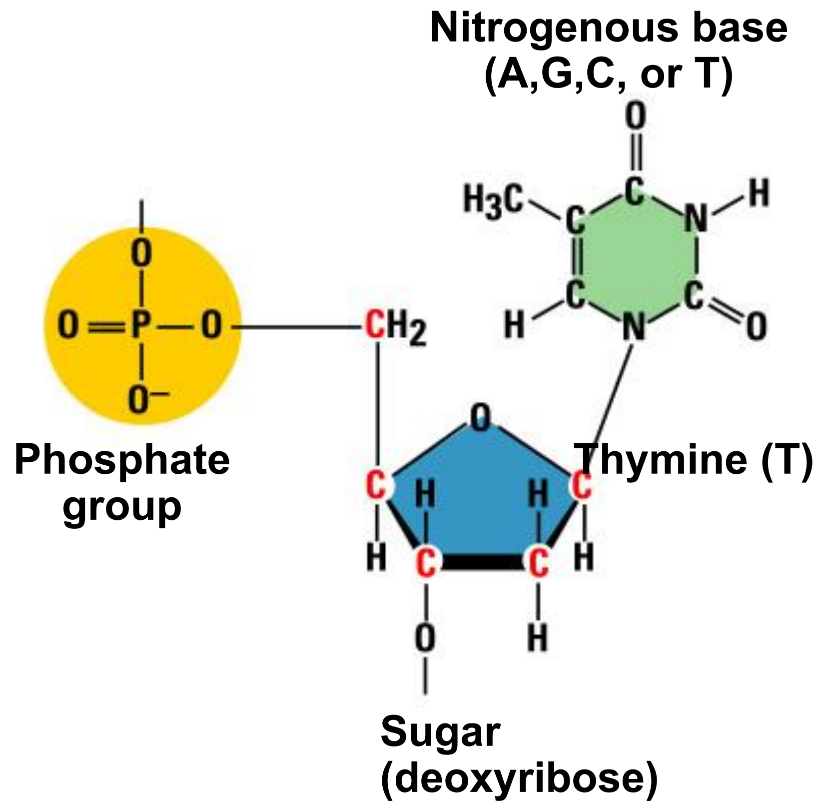
- Nucleotides form long chains called DNA

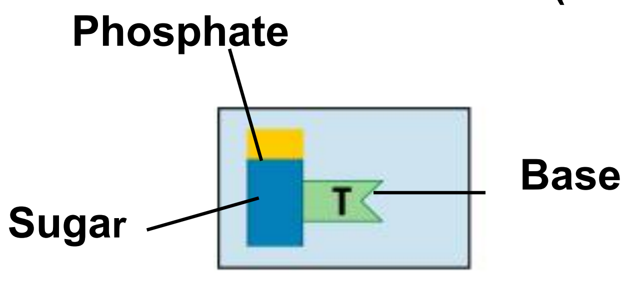
- Nucleotides are joined by sugars & phosphates on the side



Nucleic Acids

Nucleic acids are polymers of nucleotides

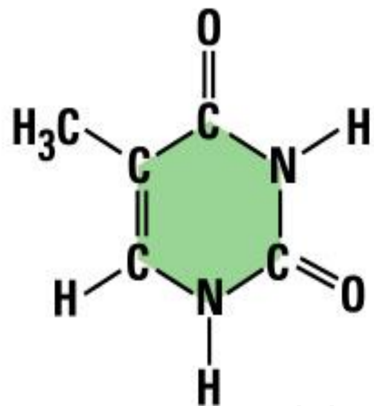


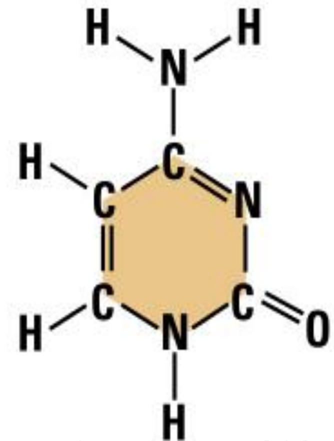


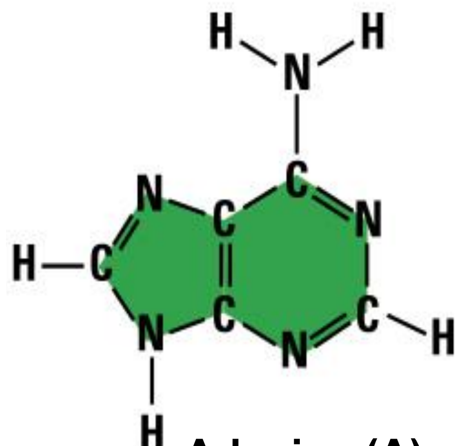
Nucleotide

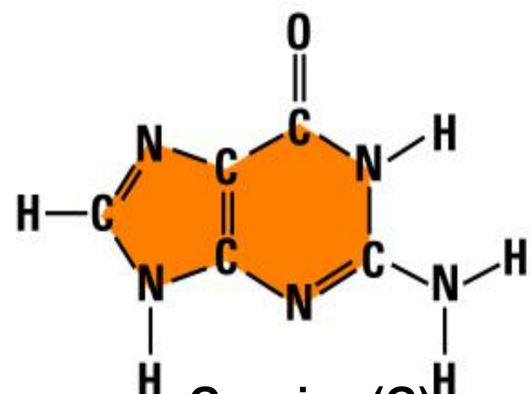
Bases

- Each DNA nucleotide has one of the following bases:
  - Adenine (A)
  - Guanine (G)
  - Thymine (T)
  - Cytosine (C)


Thymine (T)


Cytosine (C)

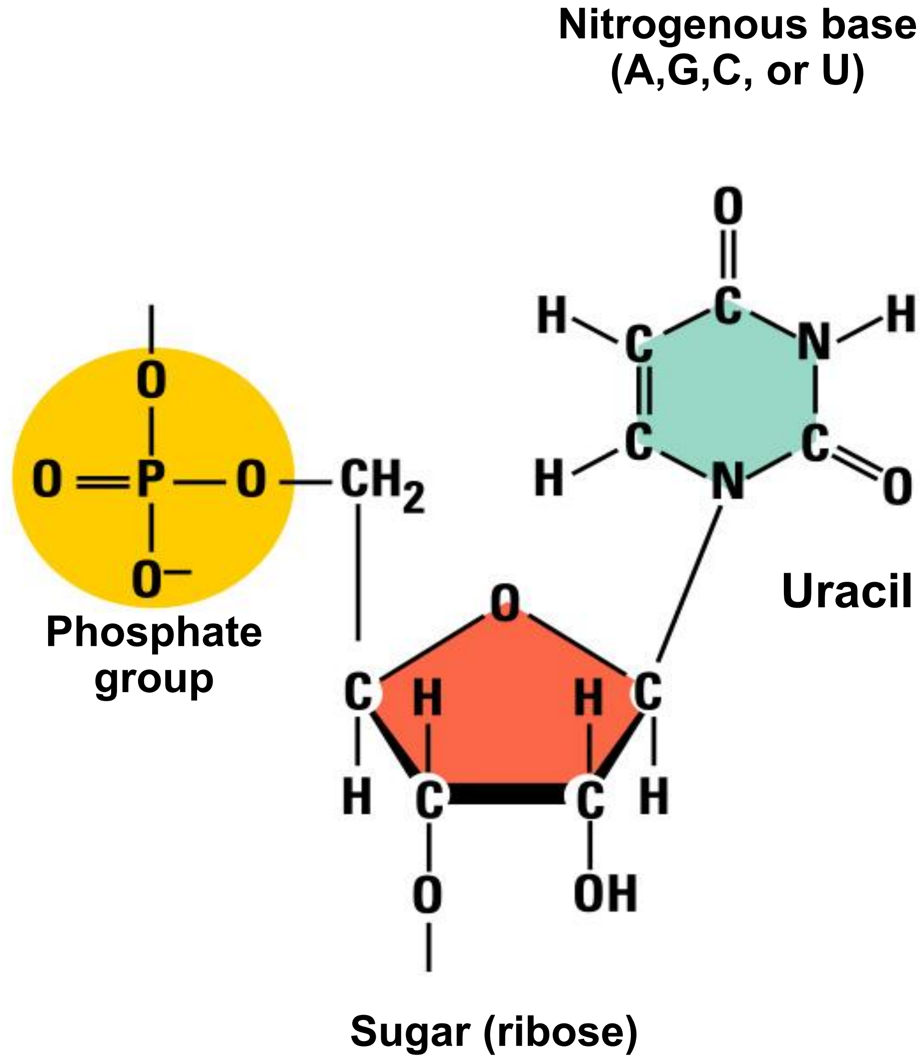

Adenine (A)


Guanine (G)

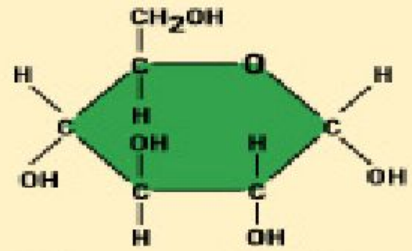
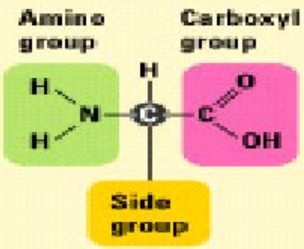
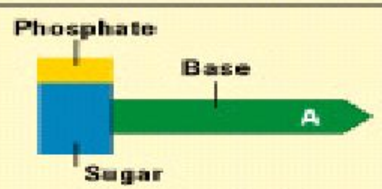
RNA – Ribonucleic Acid

- Ribose sugar has an extra –OH or hydroxyl group

- It has the base uracil (U) instead of thymine (T)



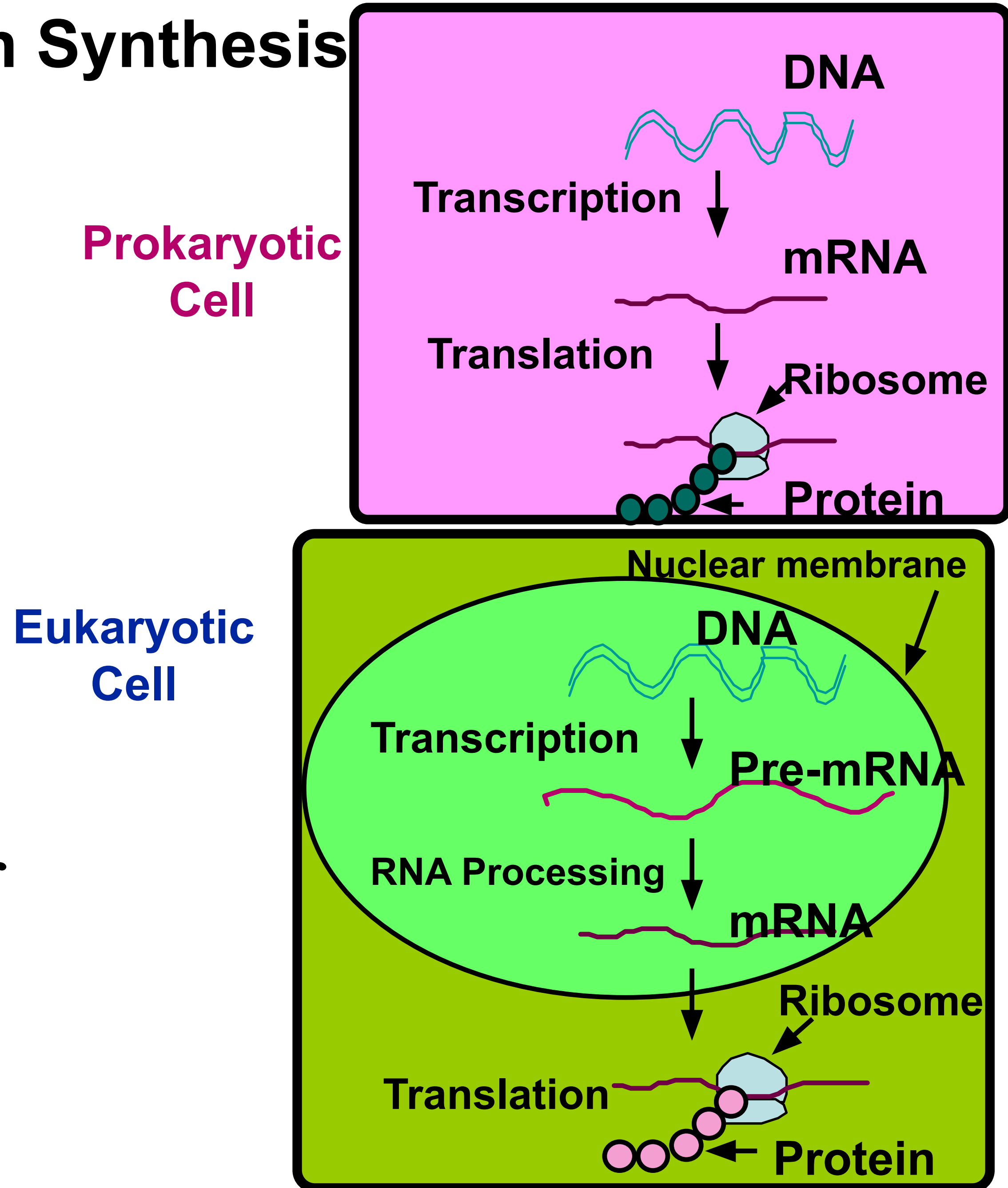
Macromolecules

Biological macromolecule	Function	Monomer	Examples
Carbohydrates	Dietary energy; storage; plant structure	 Monosaccharide	Monosaccharides: glucose, fructose. dissaccharides: lactose, sucrose. Polysaccharides: starch, cellulose.
Lipids	Long-term energy storage (for fats); hormones (for steroids)	 Components of a fat molecule	Fats, oils, steroids
Proteins	Enzymes, structure, storage, contraction, transport, etc.	 Amino acid	Lactase (an enzyme), hemoglobin
Nucleic acids	Information storage	 Nucleotide	DNA, RNA

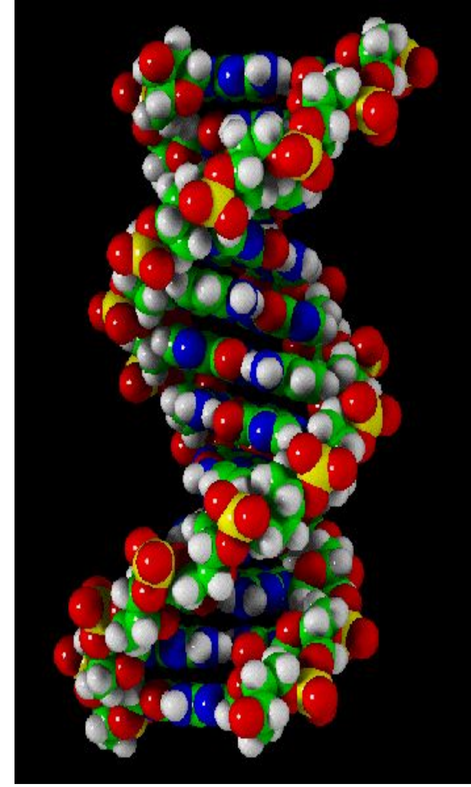
Protein Synthesis

Protein Synthesis

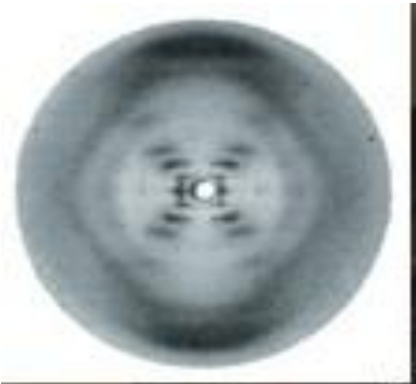
- The production (synthesis) of polypeptide chains (proteins)
- Two phases: Transcription & Translation
- mRNA must be processed before it leaves the nucleus of eukaryotic cells



Discovery of DNA structure



- Walter Sutton discovered chromosomes were made of DNA and Protein
- However, scientists were NOT sure which one (protein or DNA) was the actual genetic material of the cell
- Frederick Griffith in 1928 showed the DNA was the cell's genetic material
- Rosalind Franklin took diffraction x-ray photographs of DNA crystals
- Watson & Crick in the 1950's built
  of DNA



Rosalind Franklin

Discovery of DNA Structure

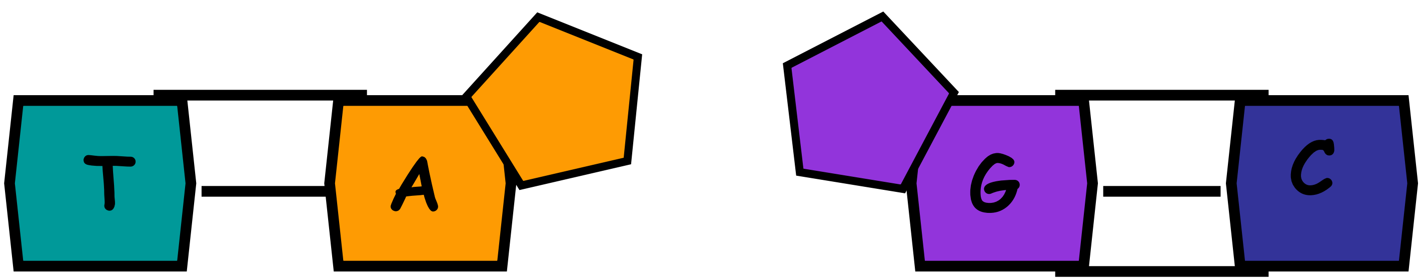
- Erwin Chargaff showed the amounts of the four bases on DNA (A,T,C,G)
- In a body or somatic cell:

  A = 30.3%

  T = 30.3%

  G = 19.5%

  C = 19.9%
- Chargaff's rule:
  - Adenine must pair with Thymine
  - Guanine must pair with Cytosine
  - The bases form weak hydrogen bonds



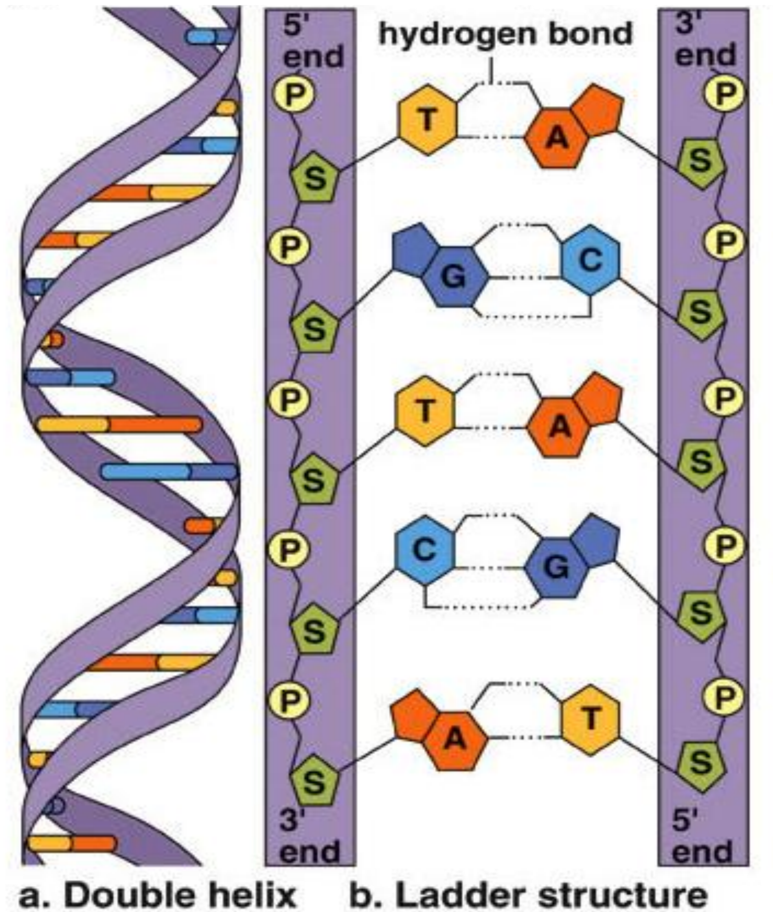
Structure of DNA

- **DNA is made of subunits called nucleotides**
- **DNA nucleotides are composed of a phosphate, deoxyribose sugar, and a nitrogen-containing base**
- **The 4 bases in DNA are: adenine (A), thymine (T), guanine (G), and cytosine (C)**
- **Purines have single rings of carbon-nitrogen (G, A)**
- **Pyrimidines have double carbon-nitrogen rings (C, T)**
- **This is called *complementary base pairing* because a purine is always paired with a pyrimidine**

5’ to 3’ Sugars

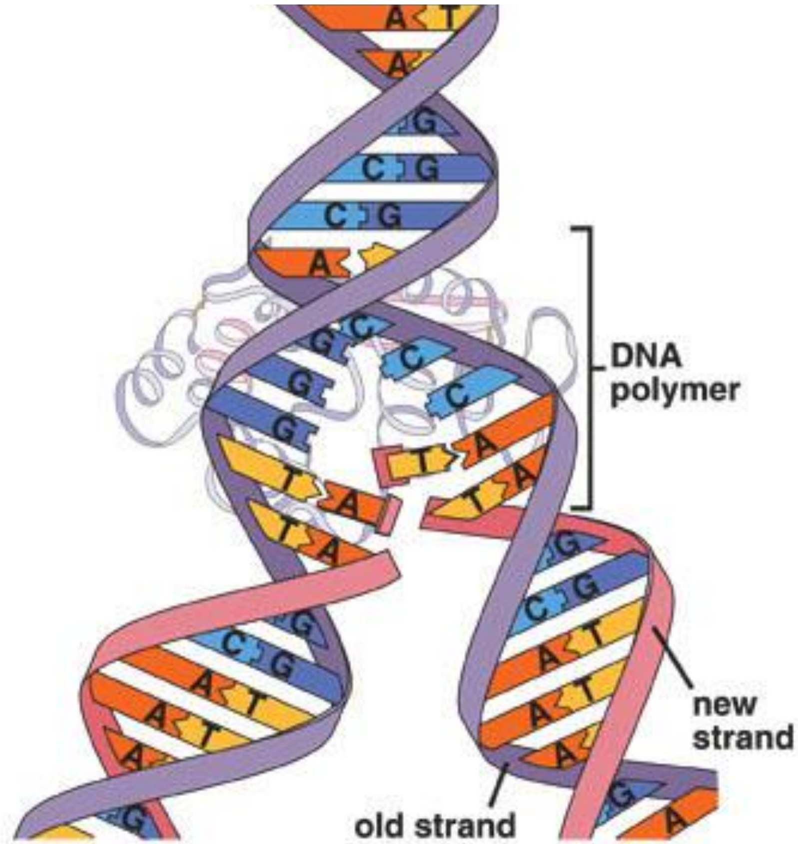
- When the DNA double helix unwinds, it resembles a ladder
- The sides of the ladder are the sugar-phosphate backbones
- The rungs of the ladder are the complementary paired bases
- The two DNA strands are anti-parallel (they run in opposite directions)

Anti-Parallel Strands of DNA



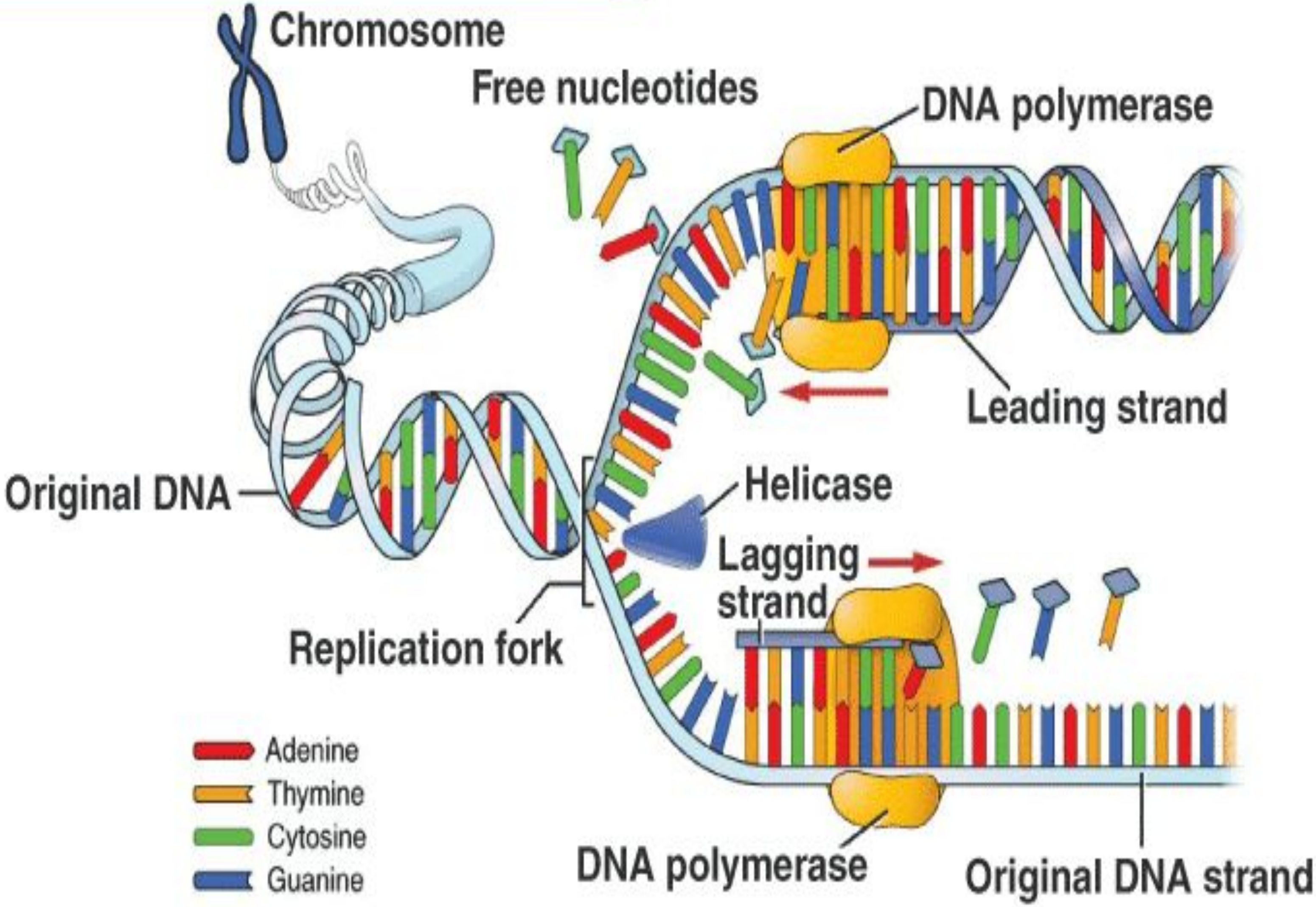
Steps in DNA Replication

- Occurs when chromosomes duplicate (make copies)
- An exact copy of the DNA is produced with the aid of the enzyme DNA polymerase
- Hydrogen bonds between bases break and enzymes "unzip" the molecule
- Each old strand of nucleotides serves as a template for each new strand
- New nucleotides move into complementary positions are joined by DNA polymerase



Two New, Identical DNA Strands Result from Replication

DNA REPLICATION



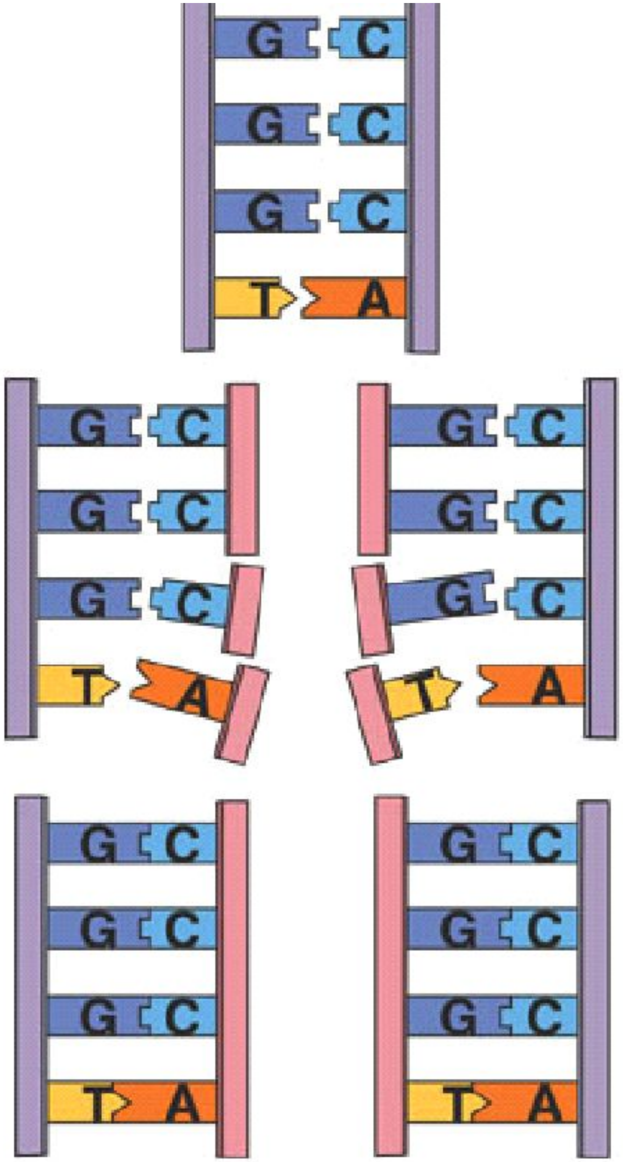
DNA Replication

- **Initiation**
- origin of replication: point where the replication originates
- Followed by unwinding of the DNA strands
- Catalyzed by enzyme: DNA helicase
- **Elongation**
- the polymerase enzymes start synthesising the complementary sequence in each of the strands
- elongation is unidirectional
- 5′ to 3′ direction
- in one strand (the template 3 ‘→5 ‘) it is continuous, hence called continuous replication while on the other strand (the template 5 ‘→3 ‘) it is discontinuous replication
- They occur as fragments called Okazaki fragments. The enzyme called DNA ligase joins them later.

DNA Replication

- **Termination**
- DNA replication fork must stop or be blocked
- involves the interaction between two components: (1) a termination site sequence in the DNA, and (2) a protein which binds to this sequence to physically stop DNA replication
- DNA replication terminus site-binding protein, or Ter protein.

Another View of Replication



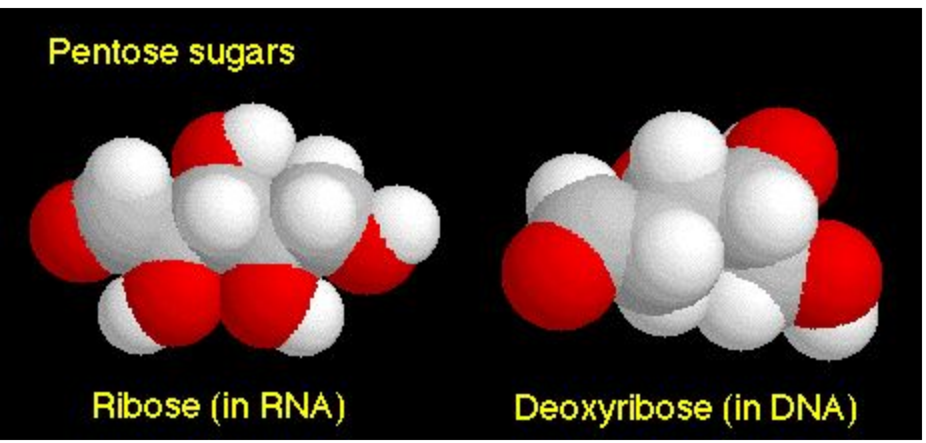
Parental DNA molecule contains so-called old strands hydrogen-bonded by complementary base pairing.

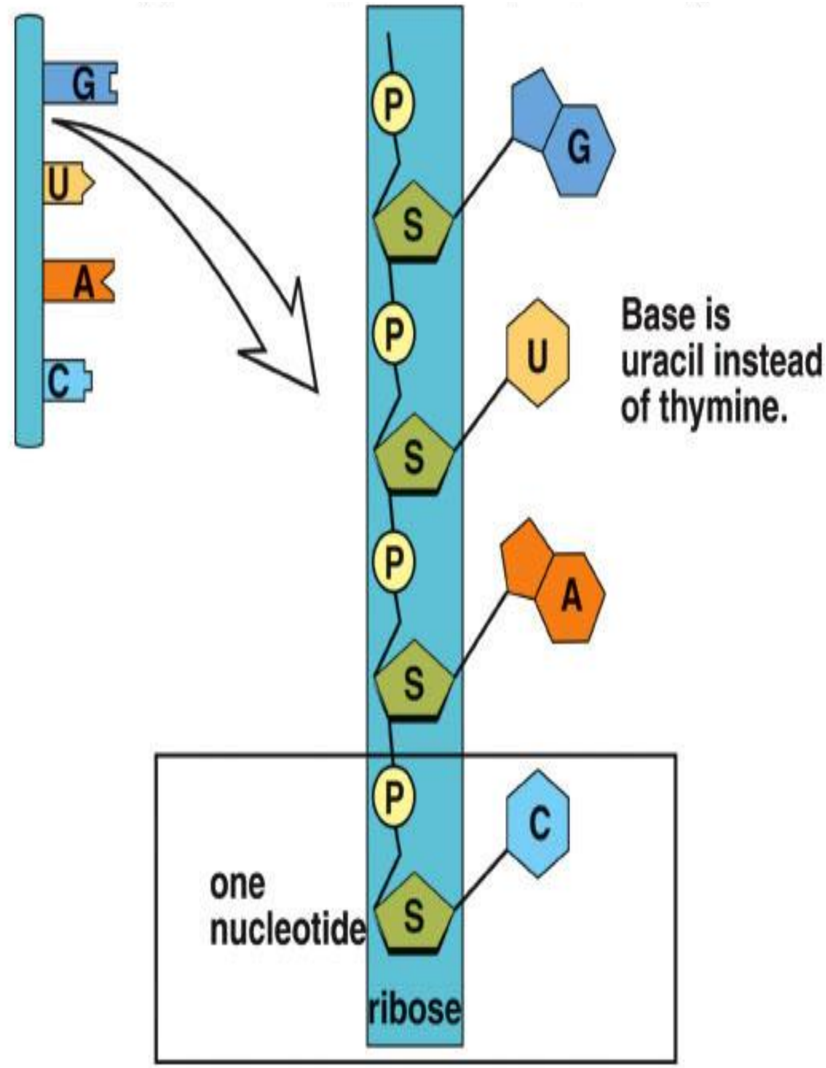
Region of replication. Parental DNA is unwound and unzipped. New nucleotides are pairing with those in old strands.

Replication is complete. Each double helix is composed of an old (parental) strand and a new (daughter) strand.

RNA Differs from DNA

1. RNA has a sugar ribose
   DNA has a sugar deoxyribose
2. RNA contains the base uracil (U)
   DNA has thymine (T)
3. RNA molecule is single-stranded
   DNA is double-stranded





Three Types of RNA

- **Messenger RNA (mRNA) carries genetic information to the ribosomes**
  (blueprint for the construction of a protein)

- **Ribosomal RNA (rRNA), along with protein, makes up the ribosomes**
  (construction site where the protein is made)

- **Transfer RNA (tRNA) transfers amino acids to the ribosomes where proteins are synthesized**
  (truck delivering the proper amino acid to the site at the right time)

Genes & Proteins

- Proteins are made of amino acids linked together by peptide bonds
- 20 different amino acids exist
- Amino acids chains are called polypeptides
- Segment of DNA that codes for the amino acid sequence in a protein are called genes

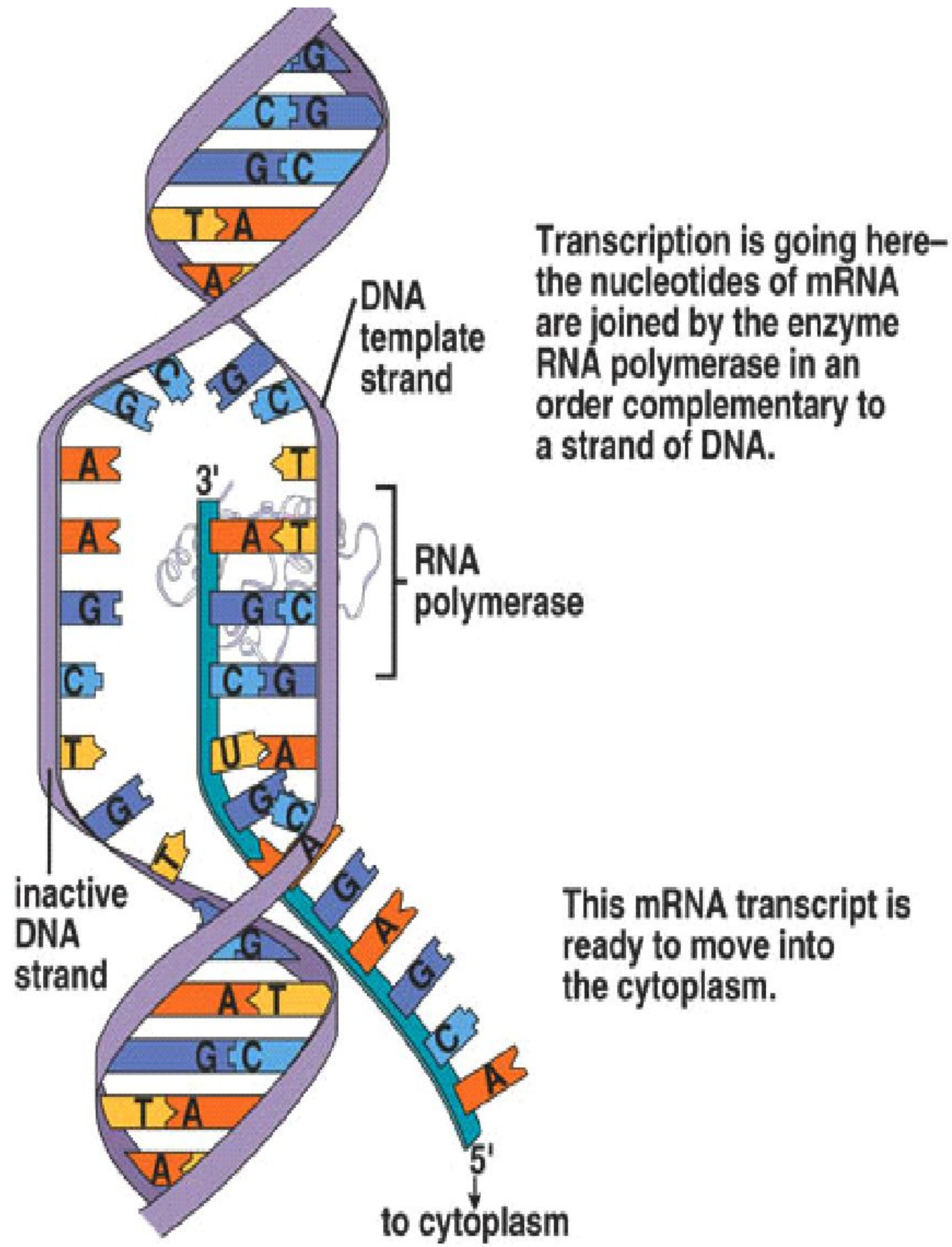
Genetic Code:

- DNA contains a triplet code
- Every three bases on DNA stands for ONE amino acid
- Each three-letter unit on mRNA is called a codon
- Most amino acids have more than one codon!
- There are 20 amino acids with a possible 64 different triplets
- The code is nearly universal among living organisms

First Base	Second Base U	Second Base C	Second Base A	Second Base G	Third Base
U	UUU phenylalanine	UCU serine	UAU tyrosine	UGU cysteine	U
U	UUC phenylalanine	UCC serine	UAC tyrosine	UGC cysteine	C
U	UUA leucine	UCA serine	UAA stop	UGA *stop*	A
U	UUG leucine	UCG serine	UAG stop	UGG tryptophan	G
C	CUU leucine	CCU proline	CAU histidine	CGU arginine	U
C	CUC leucine	CCC proline	CAC histidine	CGC arginine	C
C	CUA leucine	CCA proline	CAA glutamine	CGA arginine	A
C	CUG leucine	CCG proline	CAG glutamine	CGG arginine	G
A	AUU isoleucine	ACU threonine	AAU asparagine	AGU serine	U
A	AUC isoleucine	ACC threonine	AAC asparagine	AGC serine	C
A	AUA isoleucine	ACA threonine	AAA lysine	AGA arginine	A
A	AUG (*start*) methionine	ACG threonine	AAG lysine	AGG arginine	G
G	GUU valine	GCU alanine	GAU aspartate	GGU glycine	U
G	GUC valine	GCC alanine	GAC aspartate	GGC glycine	C
G	GUA valine	GCA alanine	GAA glutamate	GGA glycine	A
G	GUG valine	GCG alanine	GAG glutamate	GGG glycine	G

Overview of Transcription

- During transcription in the nucleus, a segment of DNA unwinds and unzips, and the DNA serves as a template for mRNA formation
- RNA polymerase joins the RNA nucleotides so that the codons in mRNA are complementary to the triplet code in DNA
- The transfer of information in the nucleus from a DNA molecule to an RNA molecule
- Only 1 DNA strand serves as the template
- Starts at promoter DNA (TATA box)
- Ends at terminator DNA (stop)
- When complete, pre-RNA molecule is released



Transcription

Initiation

- RNA polymerase binds to a sequence of DNA called the **promoter**, found near the beginning of a gene.
- Once bound, RNA polymerase separates the DNA strands, providing the single-stranded template needed for transcription.

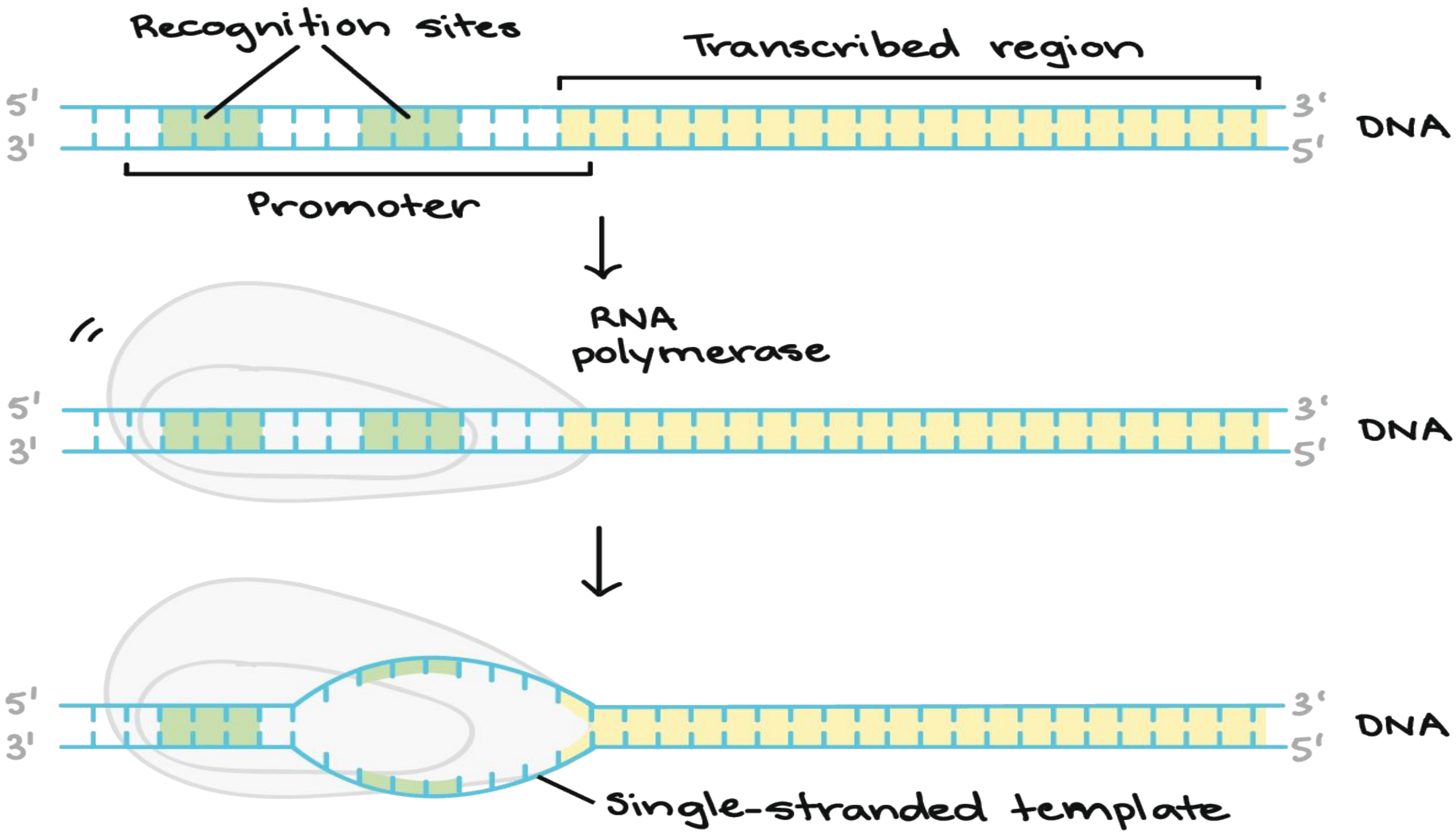
Elongation.

- One strand of DNA, the **template strand**, acts as a template for RNA polymerase.
- As it "reads" this template one base at a time, the polymerase builds an RNA molecule out of complementary nucleotides, making a chain that grows from 5' to 3'.
- The RNA transcript carries the same information as the non-template (**coding**) strand of DNA, but it contains the base uracil (U) instead of thymine (T).

Termination

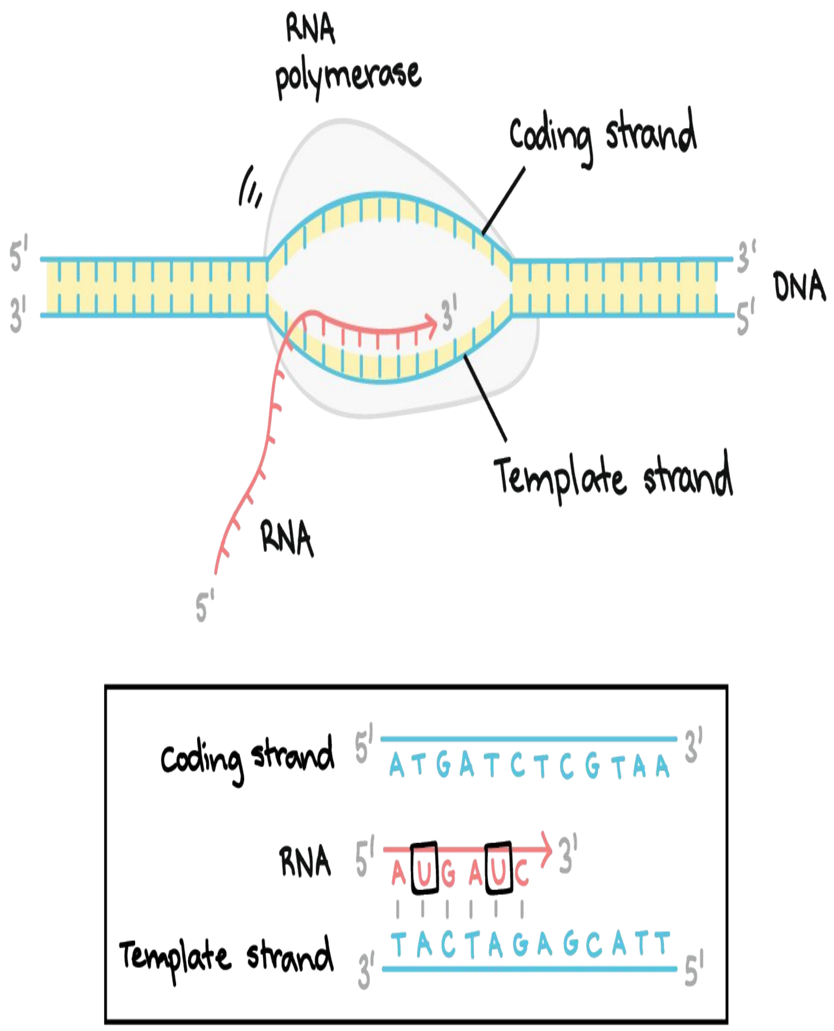
- Sequences called **terminators** signal that the RNA transcript is complete.
- Once they are transcribed, they cause the transcript to be released from the RNA polymerase.

Initiation

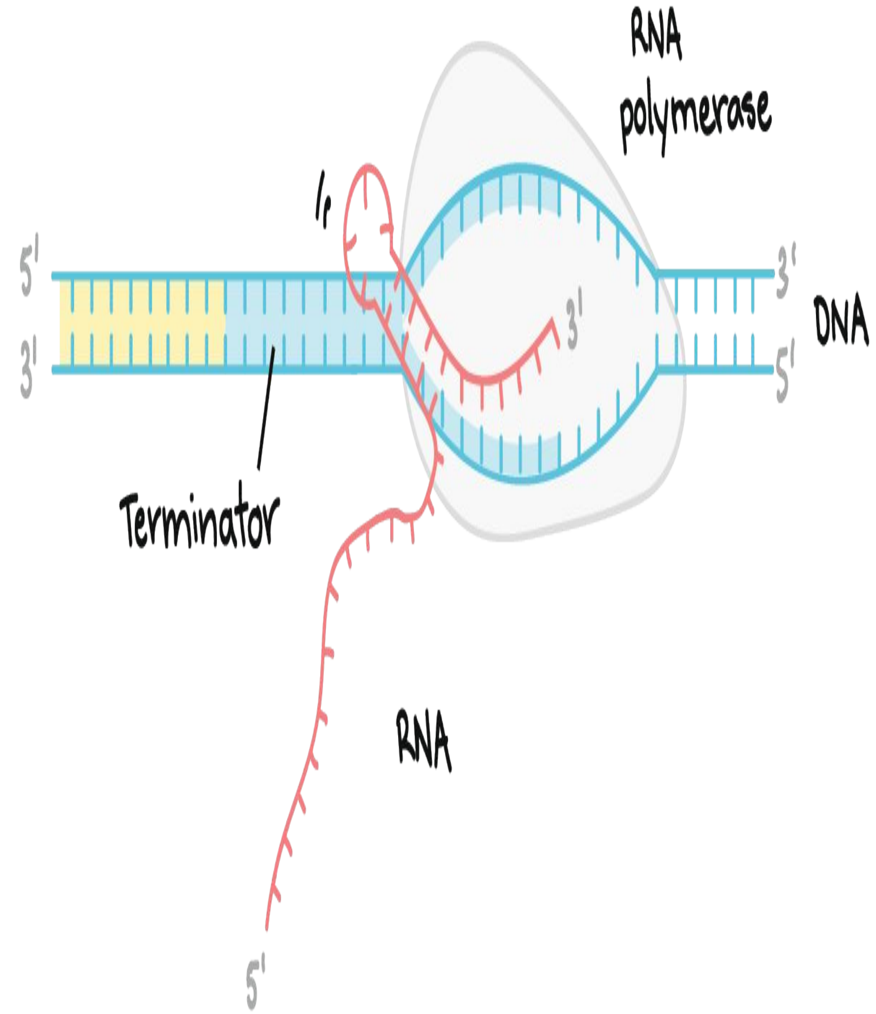


Recognition sites
Transcribed region
5'
3'
3'
5'
DNA
Promoter
RNA
polymerase
5'
3'
3'
5'
DNA
5'
3'
3'
5'
DNA
Single-stranded template

Elongation

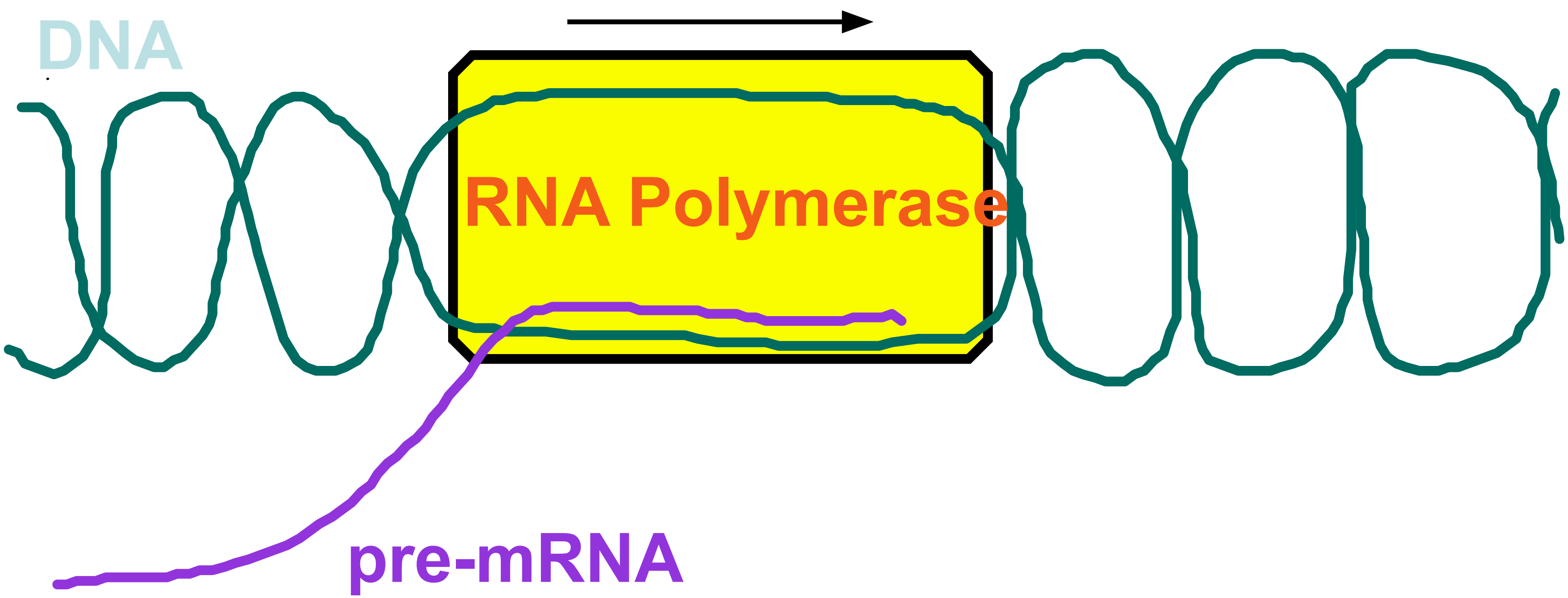


Termination



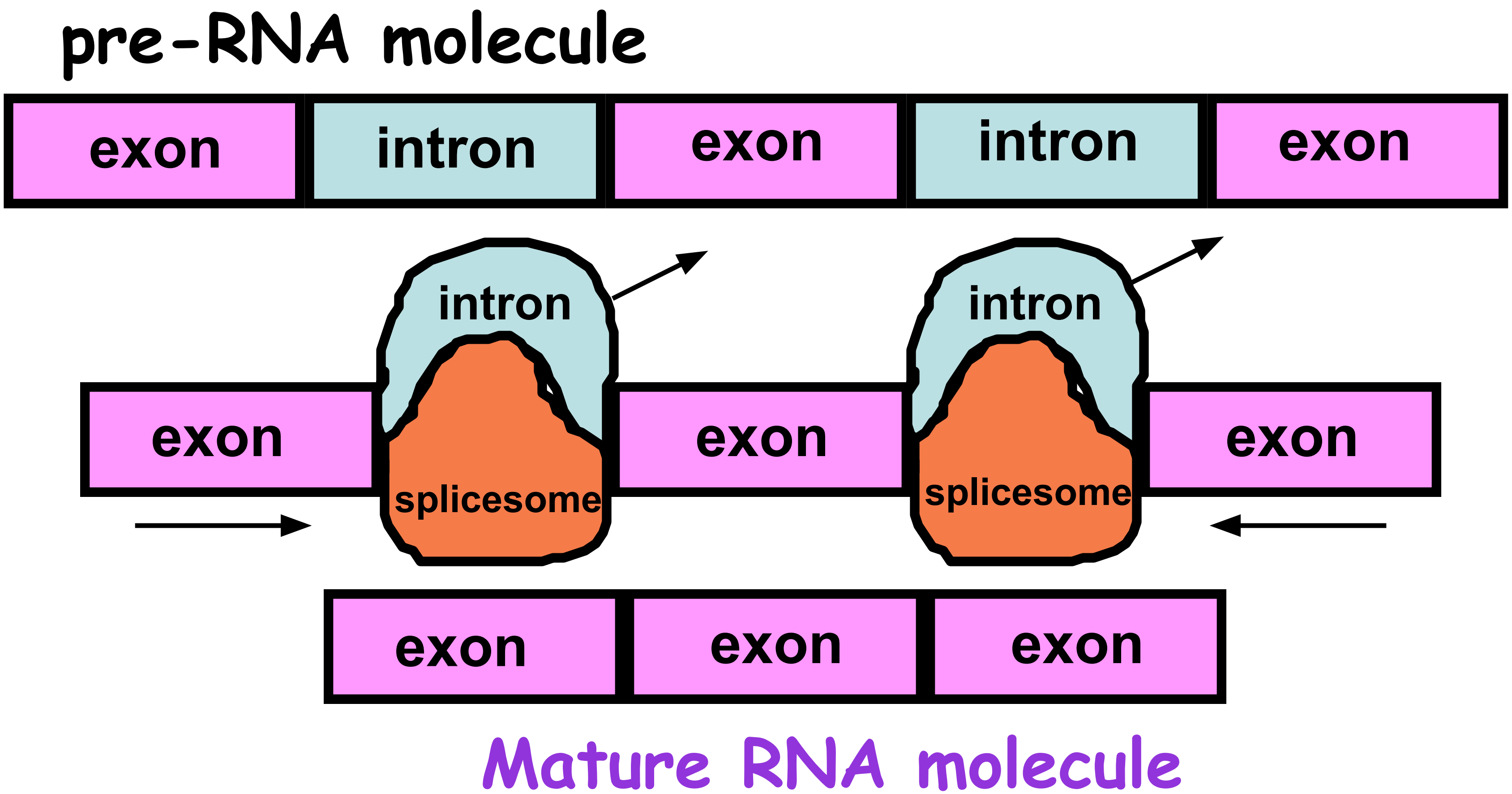
RNA Polymerase

- **Enzyme** found in the nucleus
- **Separates** the two DNA strands by **breaking the hydrogen bonds** between the bases
- **Then moves along one of the DNA strands and links RNA nucleotides together**

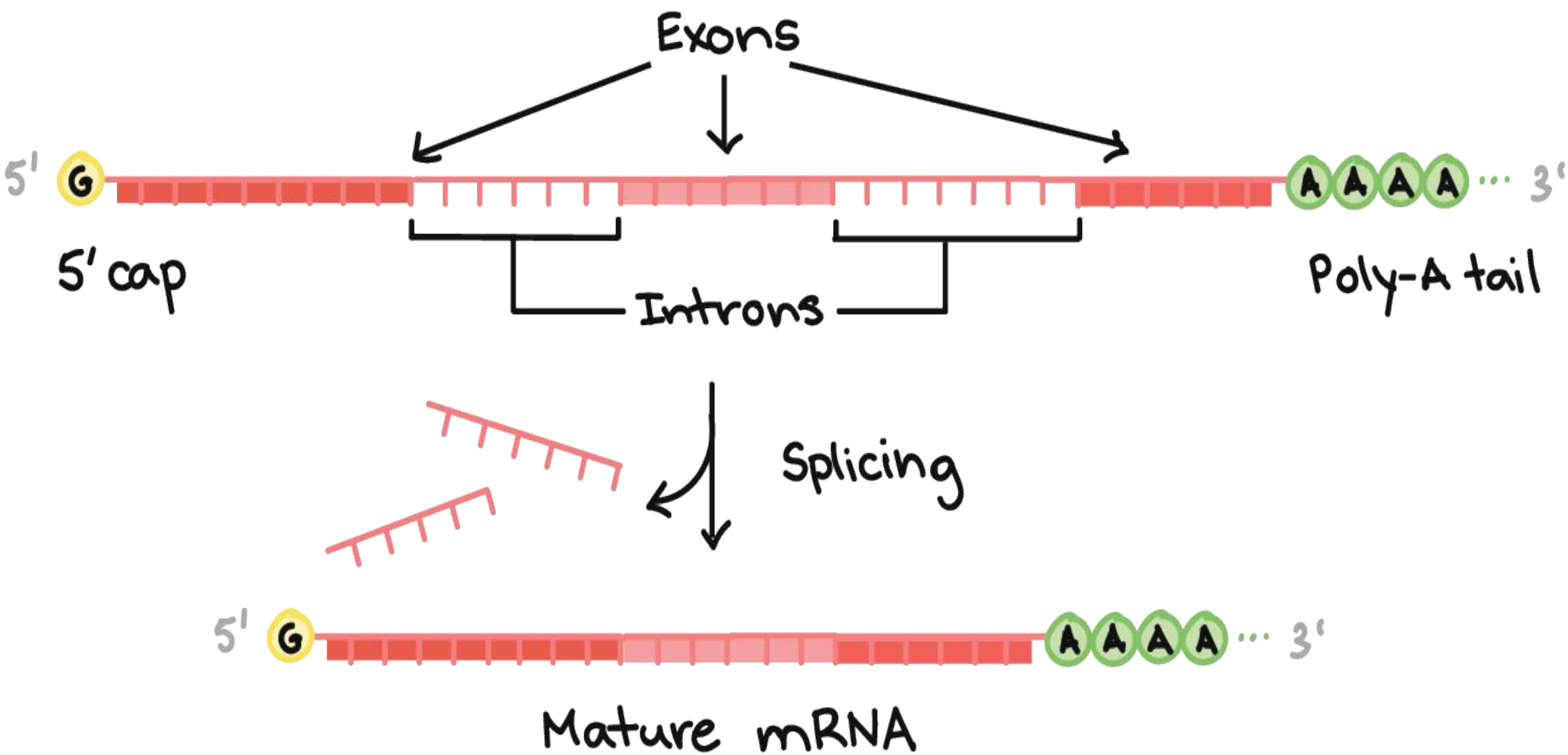


Processing Pre-mRNA

- Also occurs in the nucleus
- Pre-mRNA made up of segments called introns & exons
- Exons code for proteins, while introns do NOT!
- Introns spliced out by splicesome-enzyme and exons re-join
- End product is a mature RNA molecule that leaves the nucleus to the cytoplasm

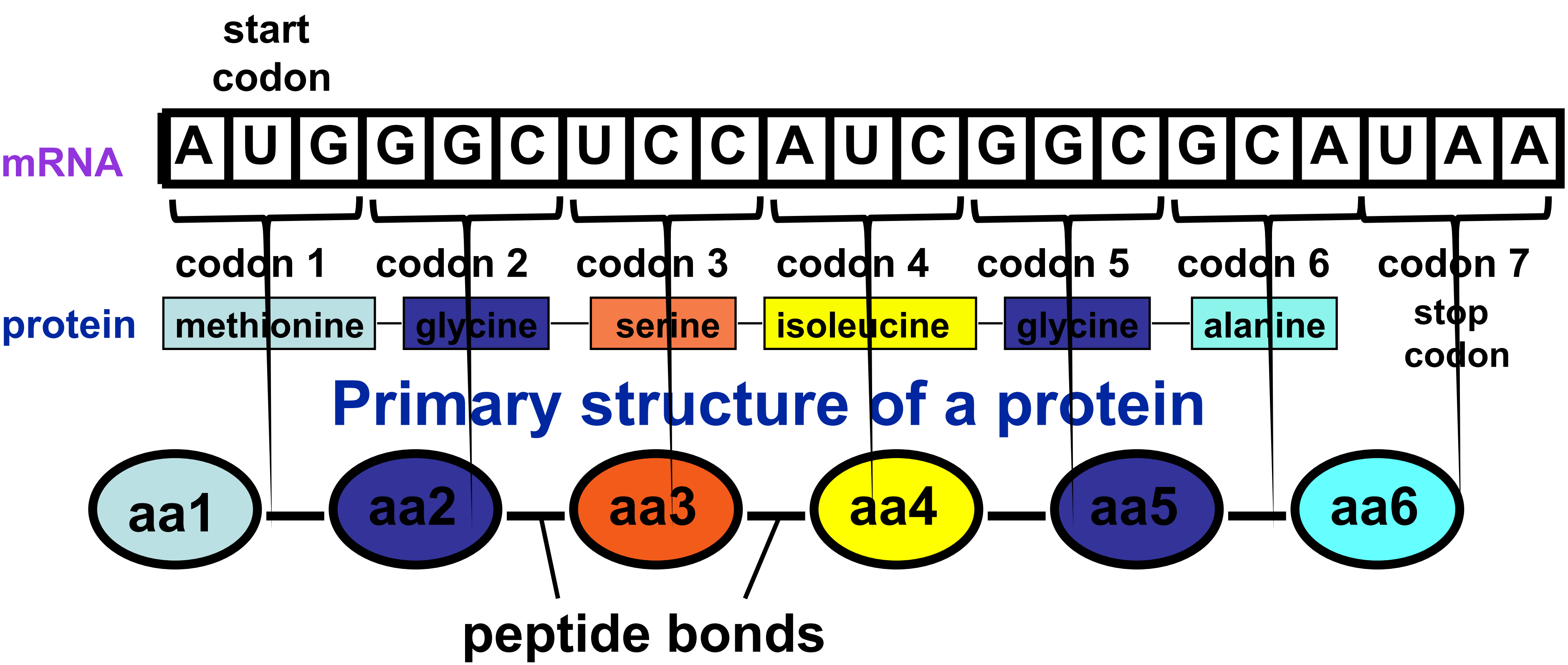


Splicing


Exons
5'
G
3'
A A A A
5' cap
Introns
Poly-A tail
Splicing
5'
G
A A A A
3'
Mature mRNA

Messenger RNA (mRNA)

- Carries the information for a specific protein
- Made up of 500 to 1000 nucleotides long
- Sequence of 3 bases called codon
- AUG – methionine or start codon
- UAA, UAG, or UGA – stop codons



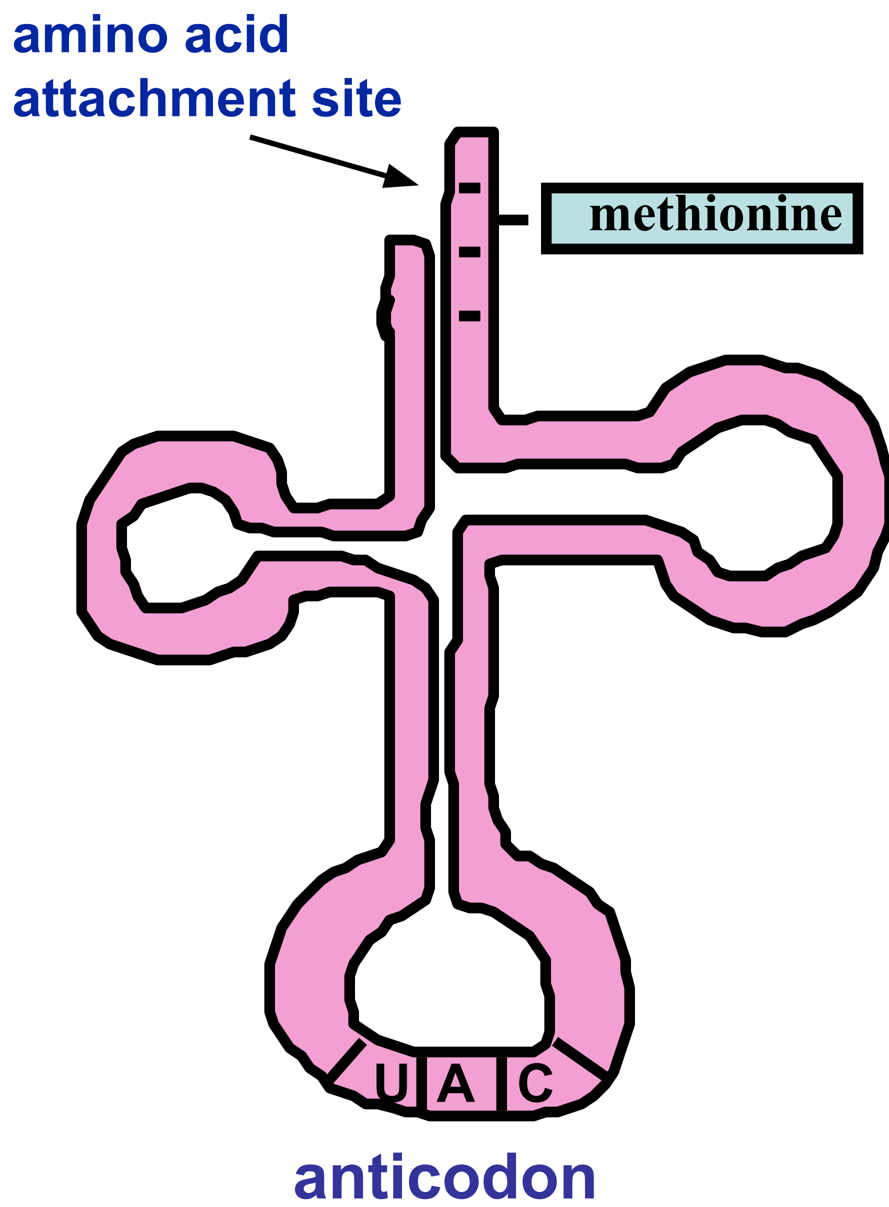
Transfer RNA (tRNA)

- Made up of 75 to 80 nucleotides long
- Clover leaf structure

- Picks up the appropriate amino acid floating in the cytoplasm
- Transports amino acids to the mRNA

- Have anticodons that are complementary to mRNA codons

- Recognizes the appropriate codons on the mRNA and bonds to them with H-bonds
- Four ATP's are required for each amino acid added to the polypeptide chain:Two to "charge" the tRNA , one to carry the charged tRNA to the ribosome and one to move the ribosome to the next codon.



Ribosomal RNA (rRNA)

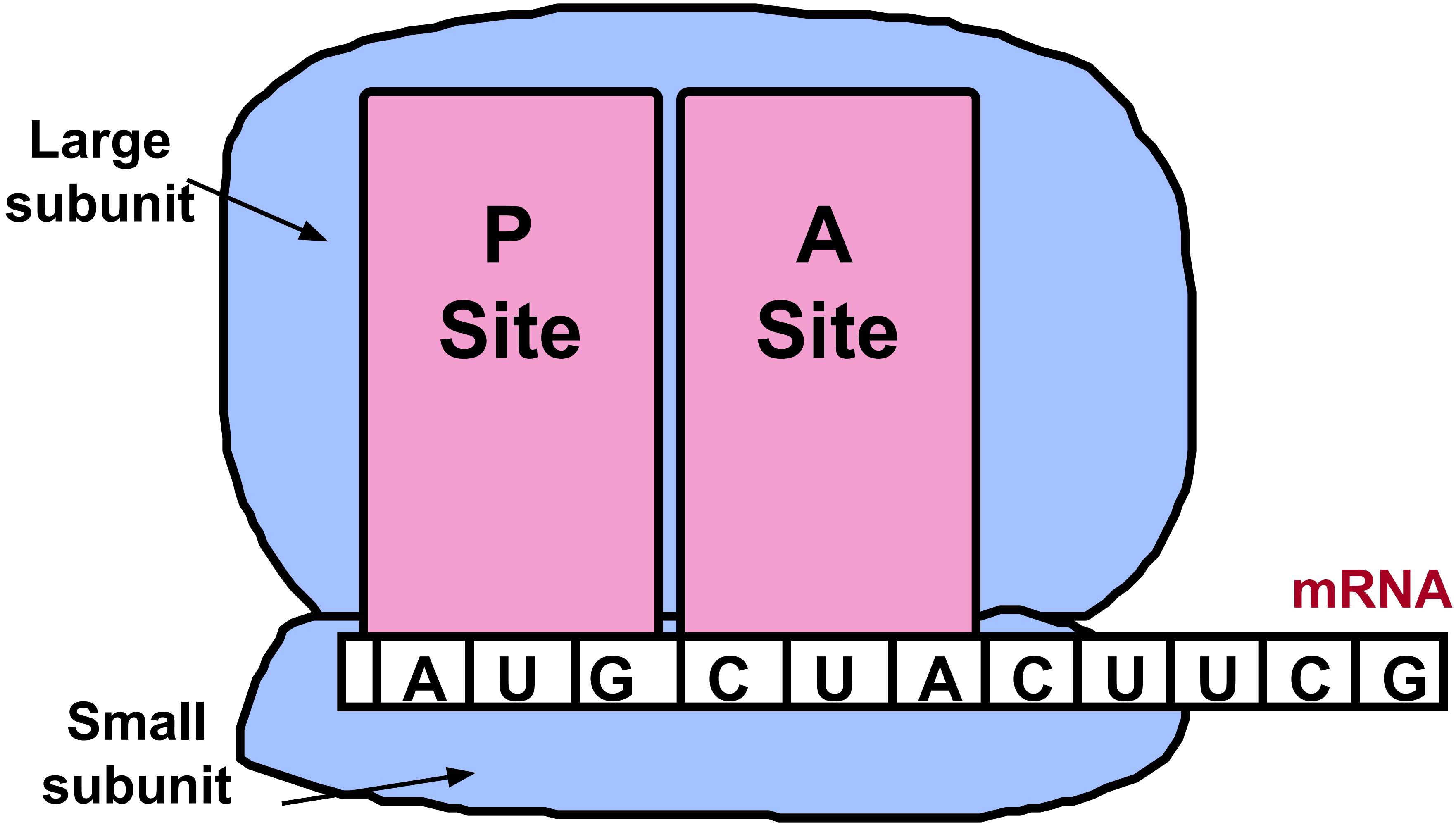
- Made up of rRNA is 100 to 3000 nucleotides long
- Made inside the nucleus of a cell
- Associates with proteins to form ribosomes

Ribosomes

- Made of a large and small subunit
- Composed of rRNA (40%) and proteins (60%)
- Have two sites for tRNA attachment --- P and A

Ribosomes

P= Peptide site
A= Amino acid site



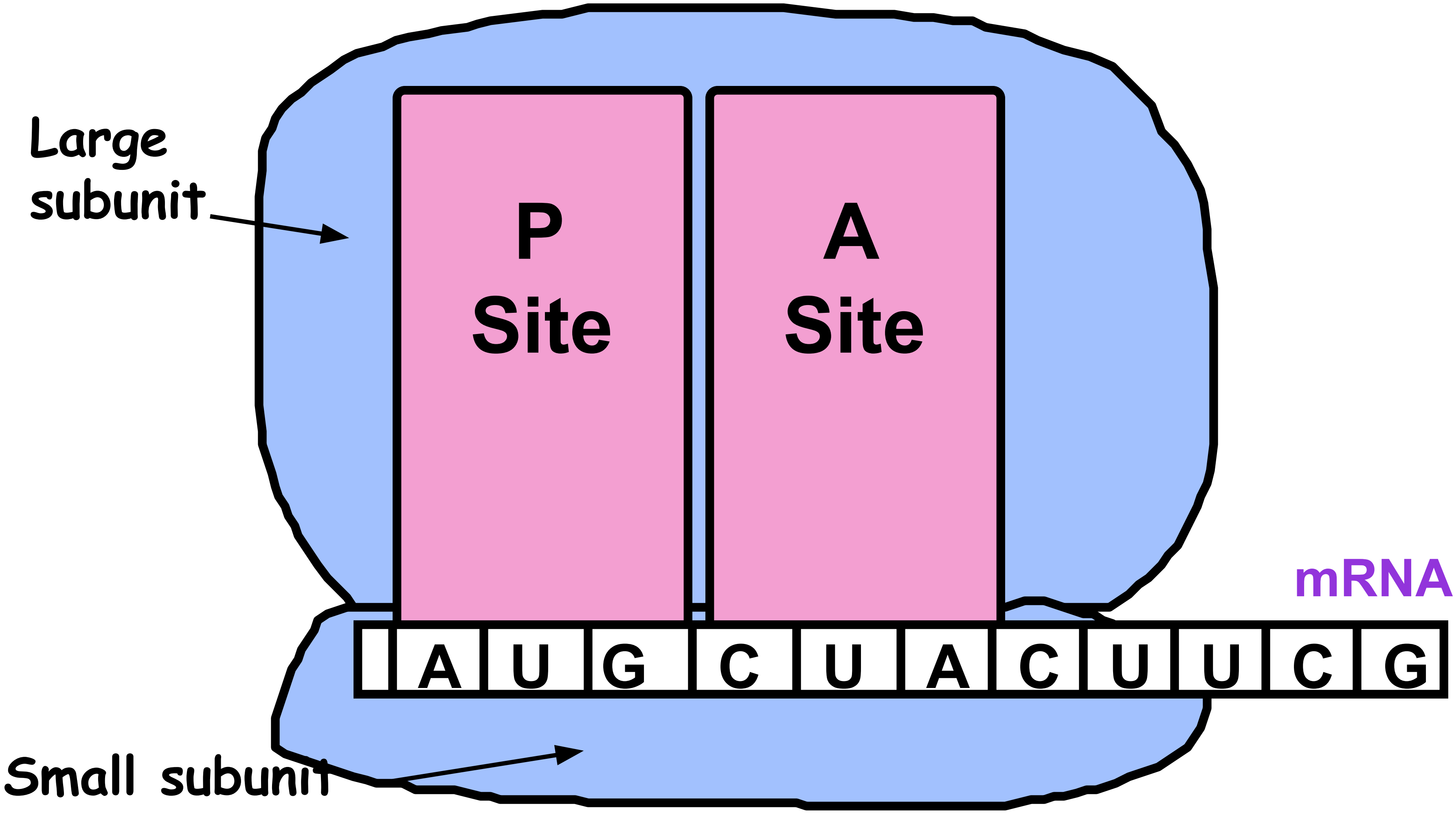
Translation

- Synthesis of proteins in the cytoplasm

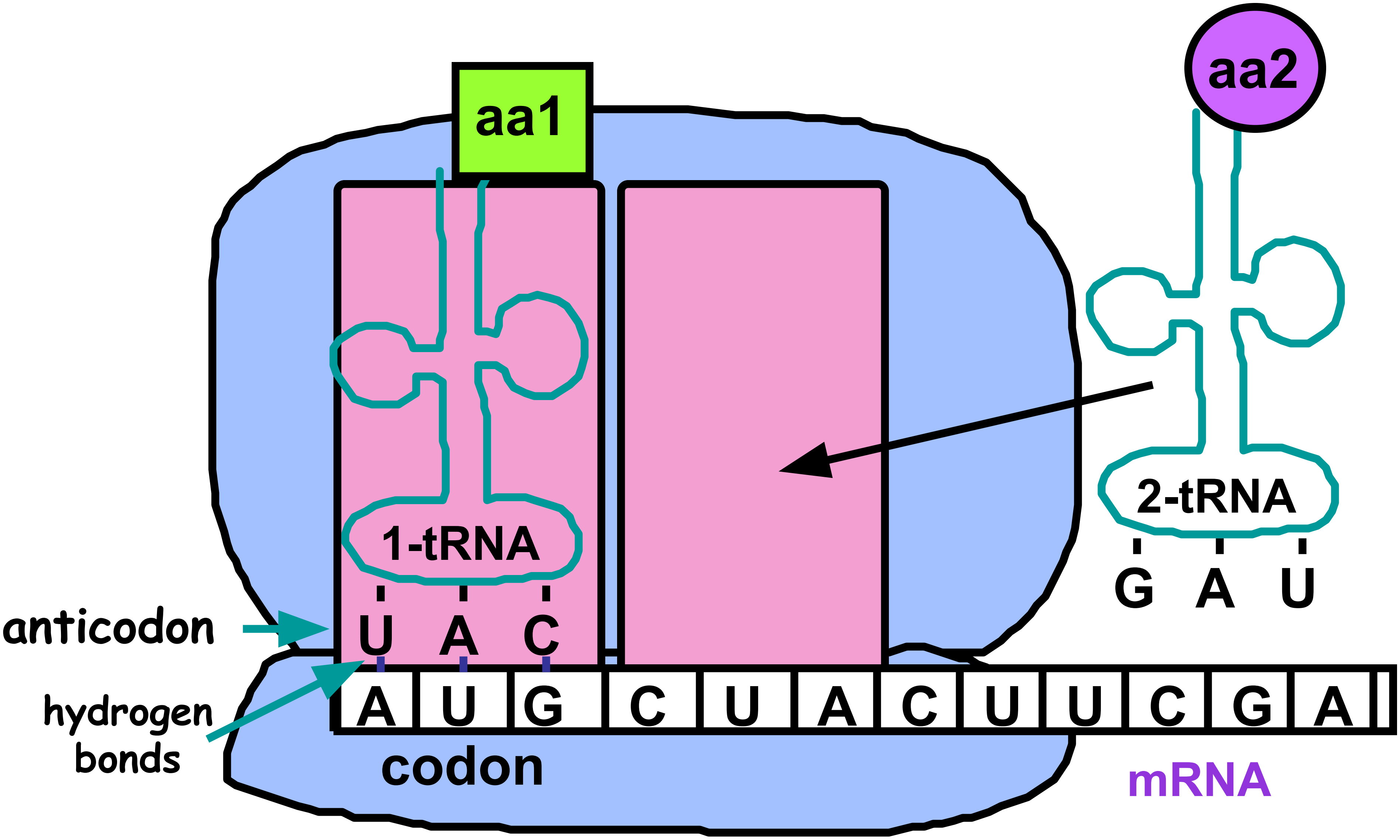
- Involves the following:
  1. mRNA (codons)
  2. tRNA (anticodons)
  3. ribosomes
  4. amino acids

- Three steps:
  1. initiation: start codon (AUG)
  2. elongation: amino acids linked
  3. termination: stop codon (UAG, UAA, or UGA).

mRNA Codons Join the Ribosome



Initiation


aa1
aa2
1-tRNA
2-tRNA
G A U
anticodon
U A C
hydrogen bonds
A U G C U A C U U C G A
codon
mRNA

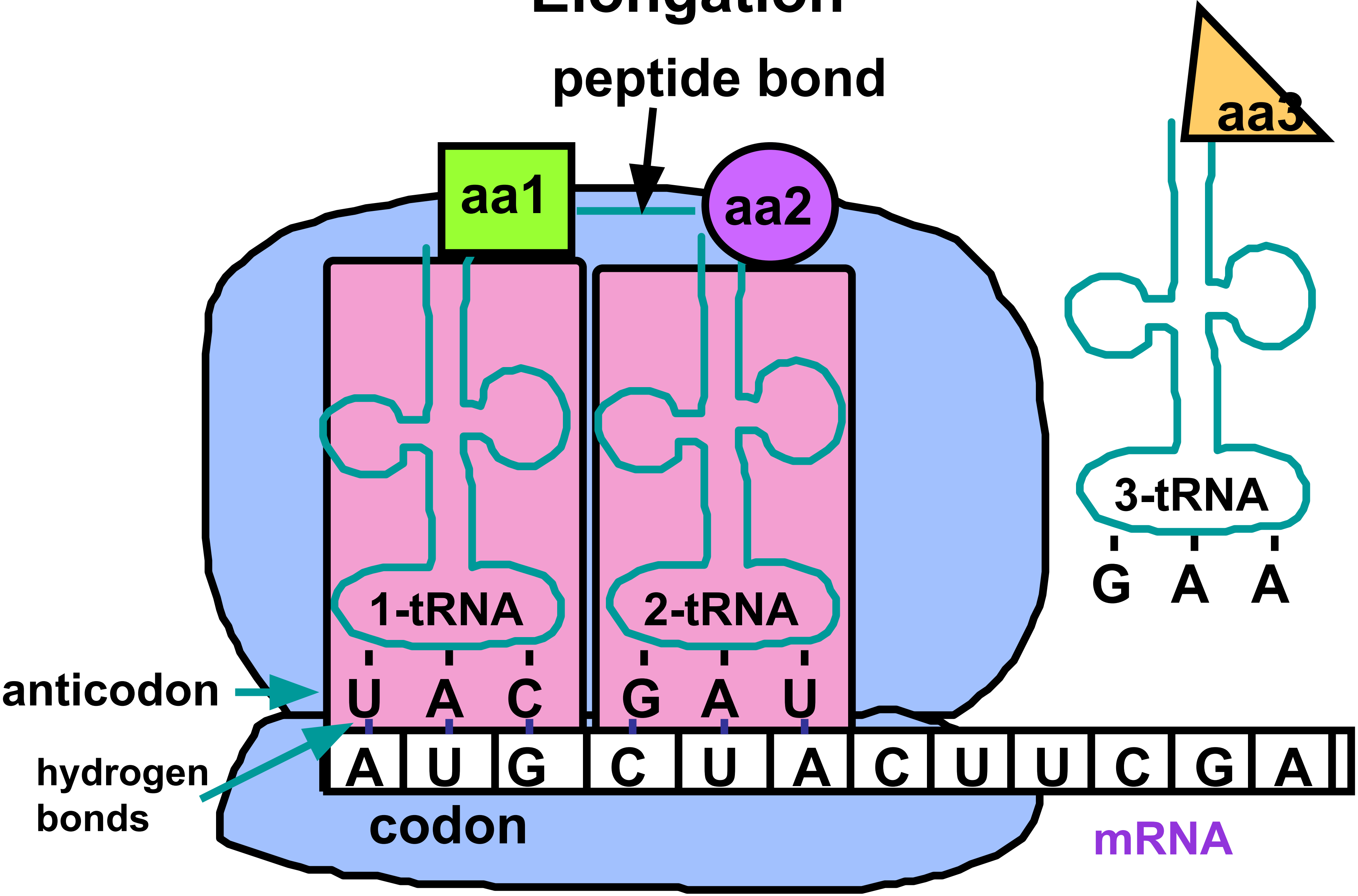
Initiation

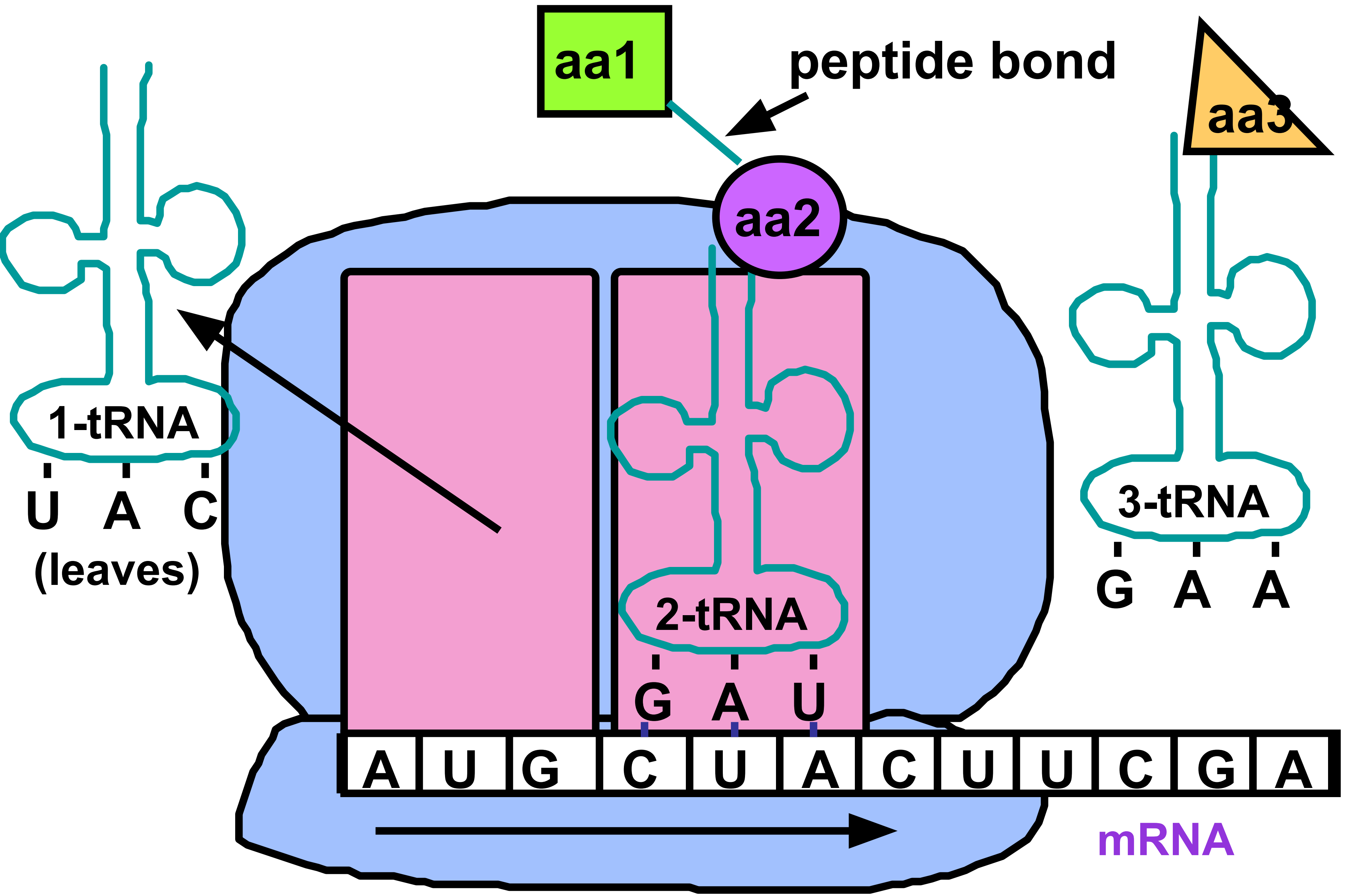
- mRNA binds to smaller unit of ribosome (30S).
- Attachment is made at the first codon of mRNA (AUG coding for amino acid methionine).
- Complex formed is known as **mRNA-30S complex**.
- tRNA having anticodon complementary to AUG (UAC) will attach to initiation codon and transports methionine to 30S subunit.
- This complex of **mRNA,tRNA and 30S** is called **pre-initiation complex.**
- The complex has two points of entry of tRNA. They are P site(peptidyl site) and A site (aminoacyl site).
- The first tRNA binds to P site.

Elongation

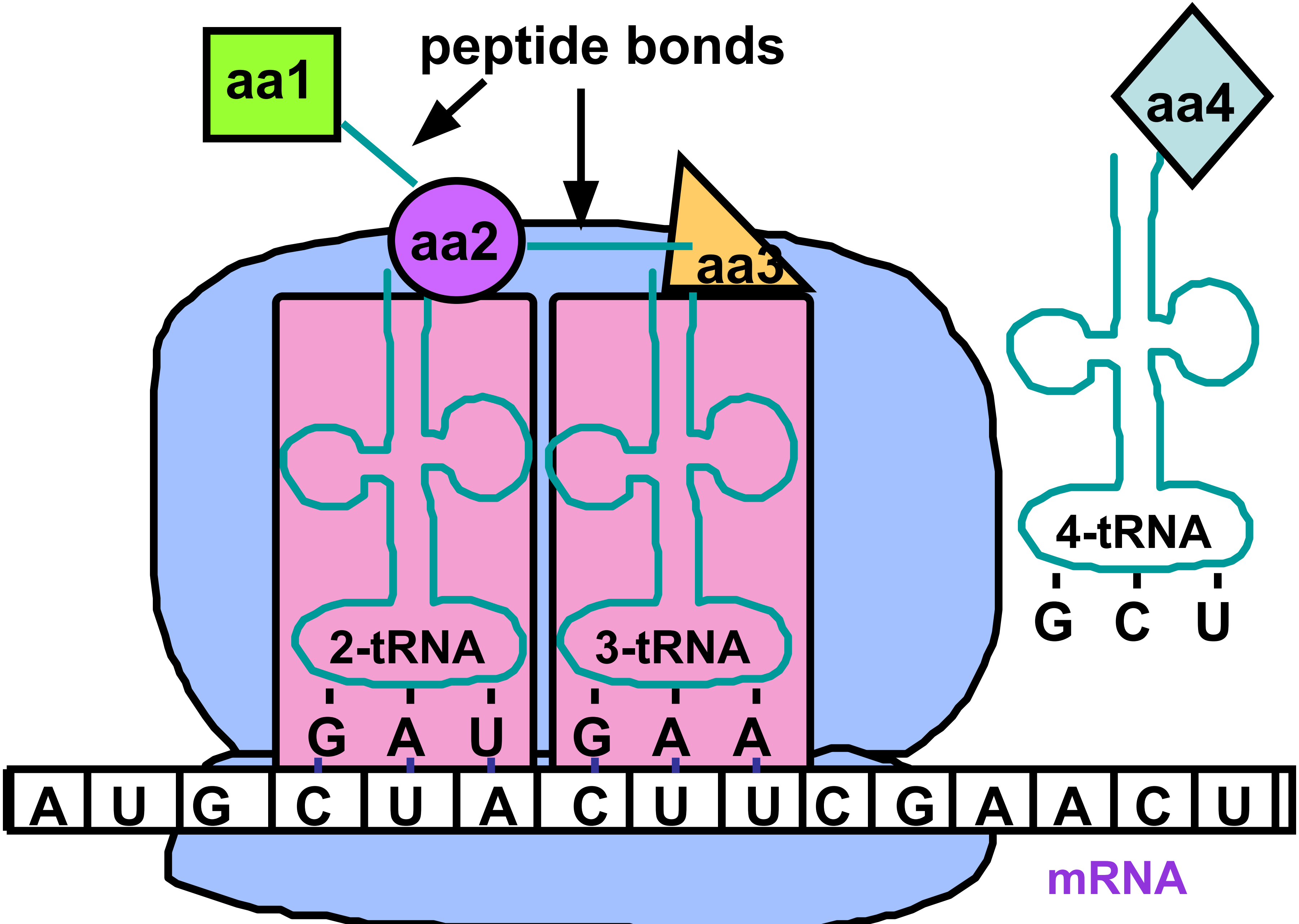
- Addition of amino acids to the first amino acids.
- The second codon in the mRNA is recognized and the corresponding enters the A site with the respective amino acid.
- tRNA with amino acid : **charged tRNA**
- Formation of peptide bond occurs between first and second amino acid.
- As the complex moves, the first tRNA leaves the complex through P site and falls off, followed by second tRNA occupying the P site and entry of new tRNA at the A site.

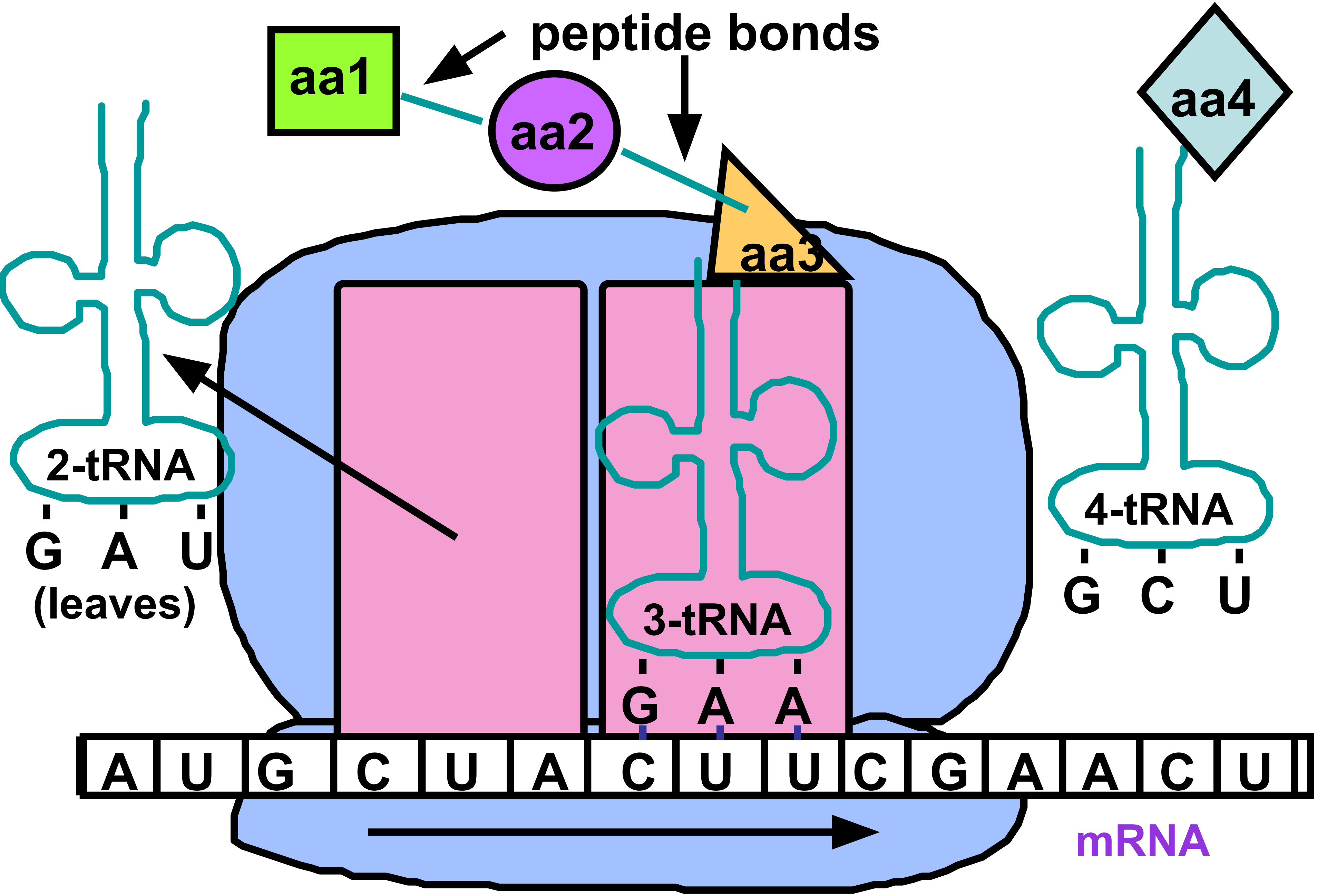
Elongation


peptide bond
aa1
aa2
aa3
1-tRNA
2-tRNA
3-tRNA
U A C
G A U
G A A
anticodon
hydrogen bonds
A U G C U A C U U C G A
codon
mRNA

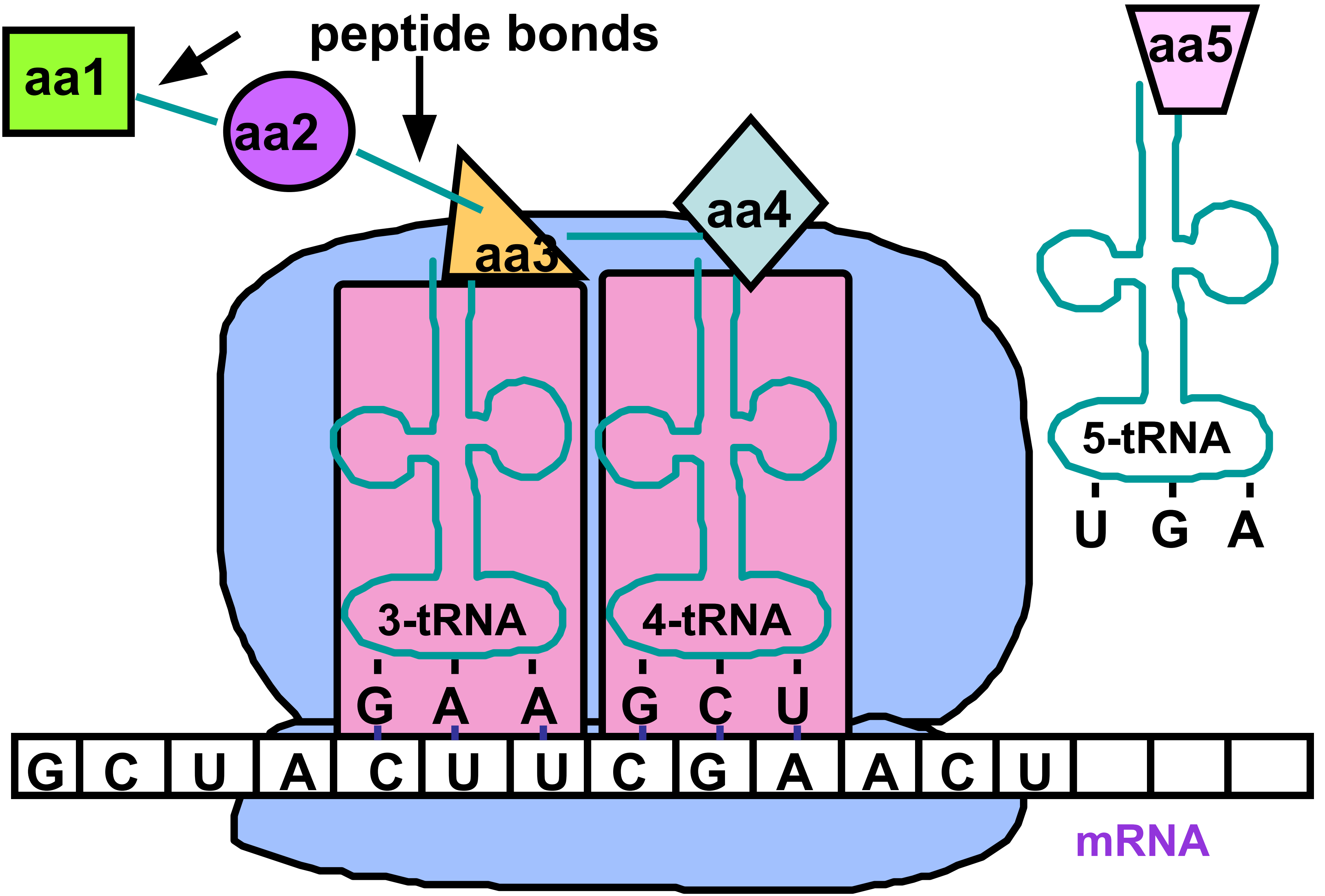


Ribosomes move over one codon

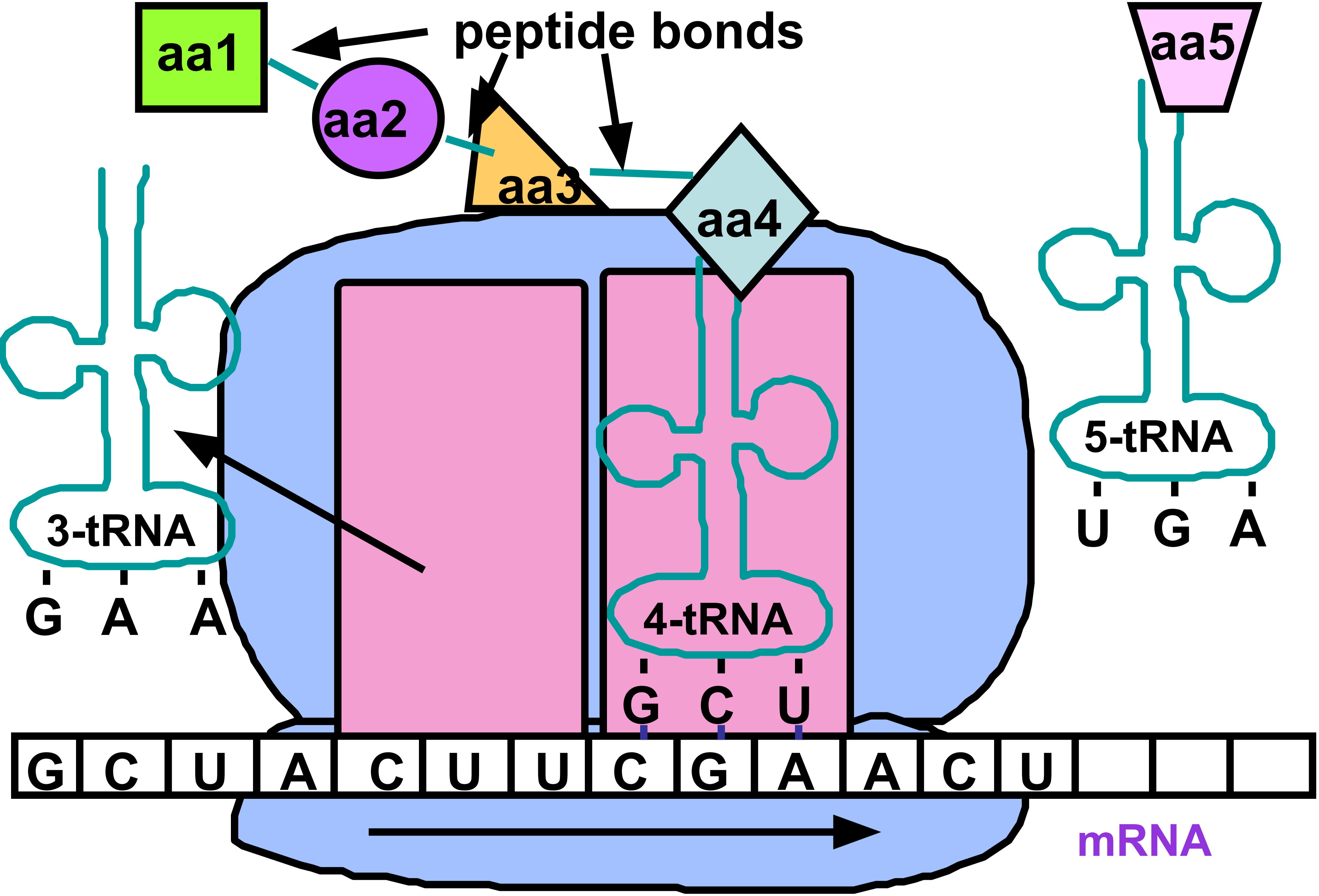

peptide bonds
aa1
aa2
aa3
aa4
2-tRNA
3-tRNA
4-tRNA
G A U
G A A
G C U
A U G C U A C U U C G A A C U
mRNA



Ribosomes move over one codon



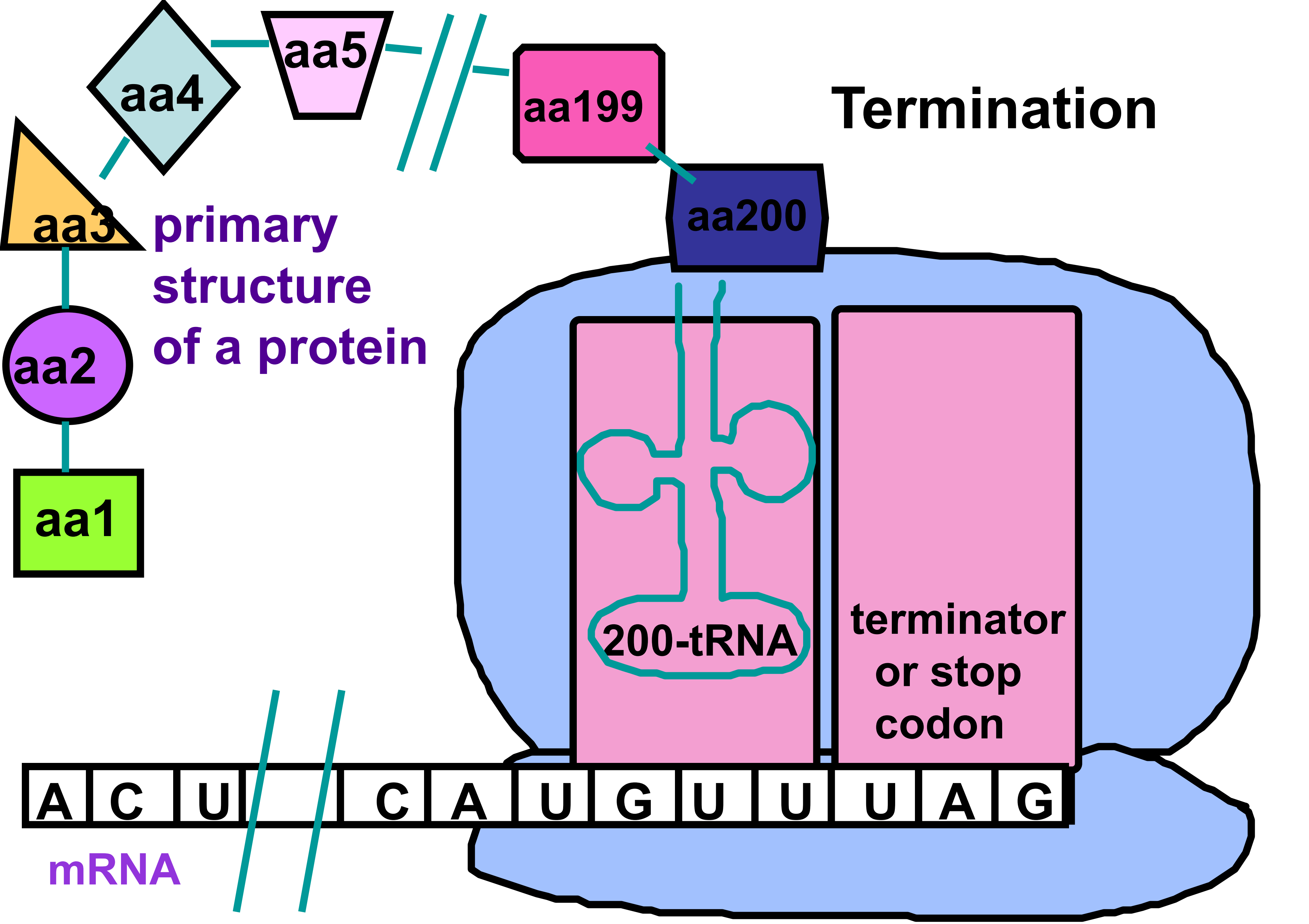
peptide bonds
aa1
aa2
aa3
aa4
aa5
3-tRNA
4-tRNA
5-tRNA
G A A
G C U
U G A
G C U A C U U C G A A C U
mRNA



Ribosomes move over one codon

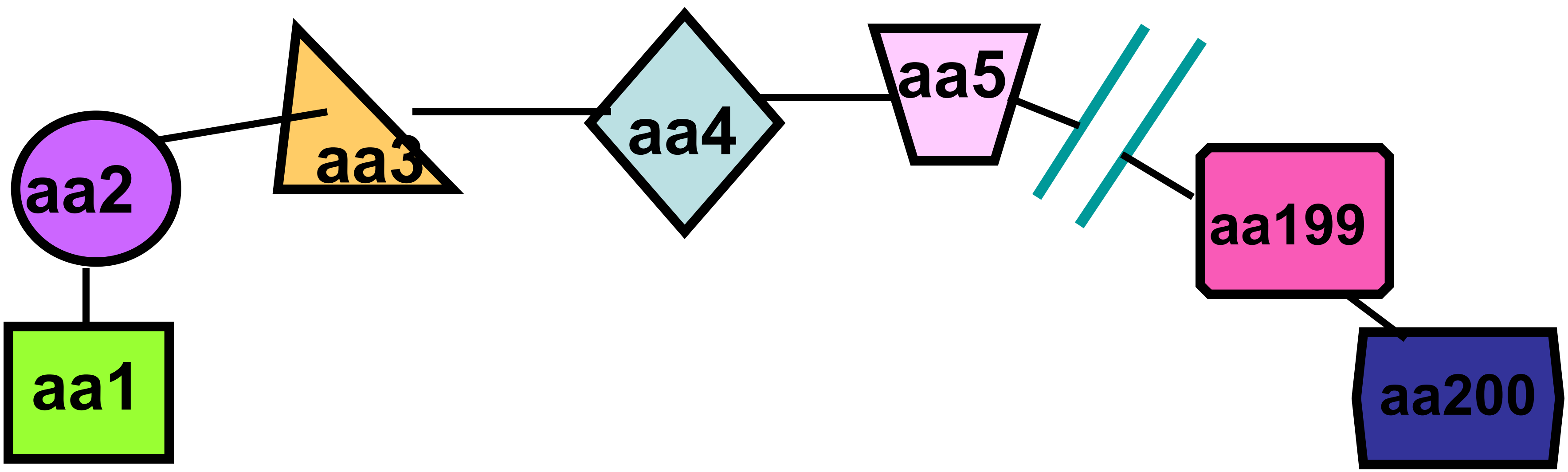
Termination

- Elongation continues till a stop codon is reached (UAA/UGA/UAG)
- Once stop codon is reached, elongation stops and the synthesized protein leaves.


Termination
aa1
aa2
aa3
aa4
aa5
aa199
aa200
primary structure of a protein
200-tRNA
terminator or stop codon
A C U C A U G U U U A G
mRNA

End Product –The Protein!

- The end products of protein synthesis is a **primary structure** of a protein
- A **sequence of amino acid** bonded together by peptide bonds



Difference between Eukaryotic & prokaryotic protein synthesis

Eukaryotes	Prokaryotes
Ribosomal subunits: 40S & 30S to form 70S	Ribosomal Units : 60S & 40S to form 80S
Multiple codons for initiation	Only one codon
Initiating amino acid is methionine	Initiating amino acid is formylated methionine
Elongation factors (EF): EF I alpha & EF I beta -gama	Elongation factors (EF): EF-Tu & EF-Ts

Stem cells and tissue engineering

Stem cell (SC)- Stem Cells are the cells which has the ability to divide for indefinite periods and which can give rise to specialized cells of various tissues of body.

Stem cells history

- **1978 - Stem cells were discovered in human cord blood**
- **1981 - First in vitro stem cell line developed from mice**
- **1998 - Researchers isolated stem cells from human embryos**
- **1999 - First Successful human transplant of insulin-making cells from cadavers**

Properties

- Two defining property-Ability to **differentiate** into other cells, ability to **self regenerate**
- It can be maintained in the *in vitro* conditions for extended period using artificial medium
- Its karyotype remains stable even after many division
- It can produce any type of adult cells of the organisms.

Sources of stem cells

1. EMBRYONIC STEM CELLS
   -Isolated from blastocyst stage of embryos
   -Pluripotent (capable of developing into almost all the cell types of the body) in nature
2. ADULT STEM CELLS
   -It is present in all the organs, but very little amount
   - They are multipotent (ability of a single stem cell to develop into more than one cell type but with ability to differentiate into a closely related family of cells).

Classification of stem cells

Unipotent - cell which can make exact copies of itself indefinitely, can differentiate, and produce same type of cells eg. Adult muscle stem cell

Totipotent- can become any cell type

eg: Morula stage of Embryos

Pluripotent- almost any kind of cell except placenta

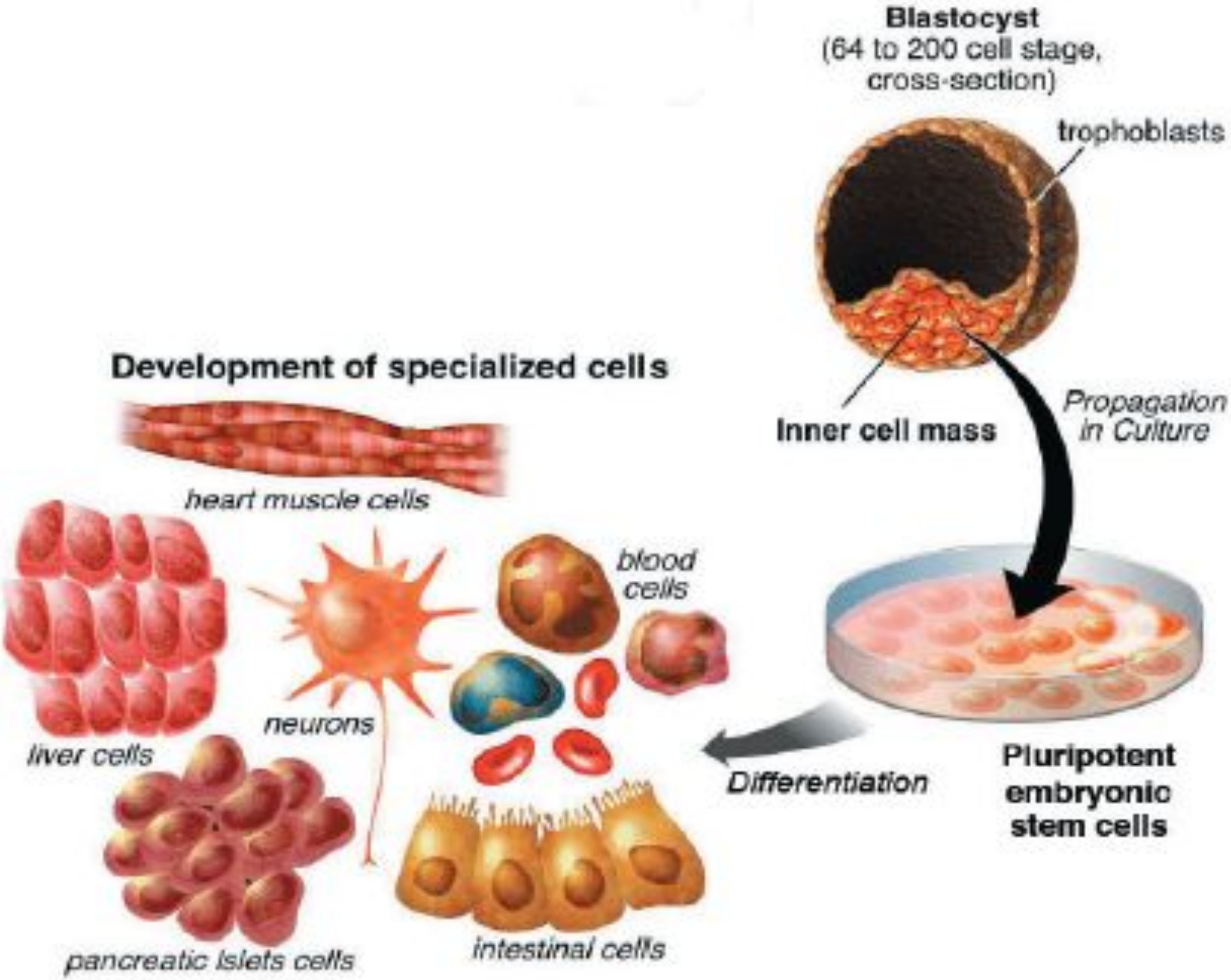
eg: Embryonic SC

Multipotent- Produces limited range of cell types

Adult SC: nerve cells, blood cells, muscle cells, bone and skin cells.

Culture of embryonic stem cells

- Collection of embryos from IVF centres
- Isolation of ICM (inner cell mass) from the blastocyst stage
- Transfer ICM to the center of culture plate containing feeder cells (UV treated mouse skin cells) and growth medium
- It can be differentiated to any cells by adding specific medium



Blastocyst
(64 to 200 cell stage,
cross-section)
trophoblasts
Inner cell mass
Propagation
in Culture
Pluripotent
embryonic
stem cells
Differentiation
Development of specialized cells
heart muscle cells
blood
cells
liver cells
neurons
pancreatic islets cells
intestinal cells

Applications

- It can be used for neural degenerative diseases (Alzheimer's and Parkinson’s disease)
- It can also used to treat diabetes by injecting *in vitro* grown pancreatic islet cells
- It is also used for treating **bone related diseases by injecting *in vitro*** developed osteocytes, chondrocytes and myocytes
- Today most of the diseases get cured through stem cell therapy eg. Muscular dystrophy, Polio.