

Course: Fundamentals of Biochemistry

Unit: 2

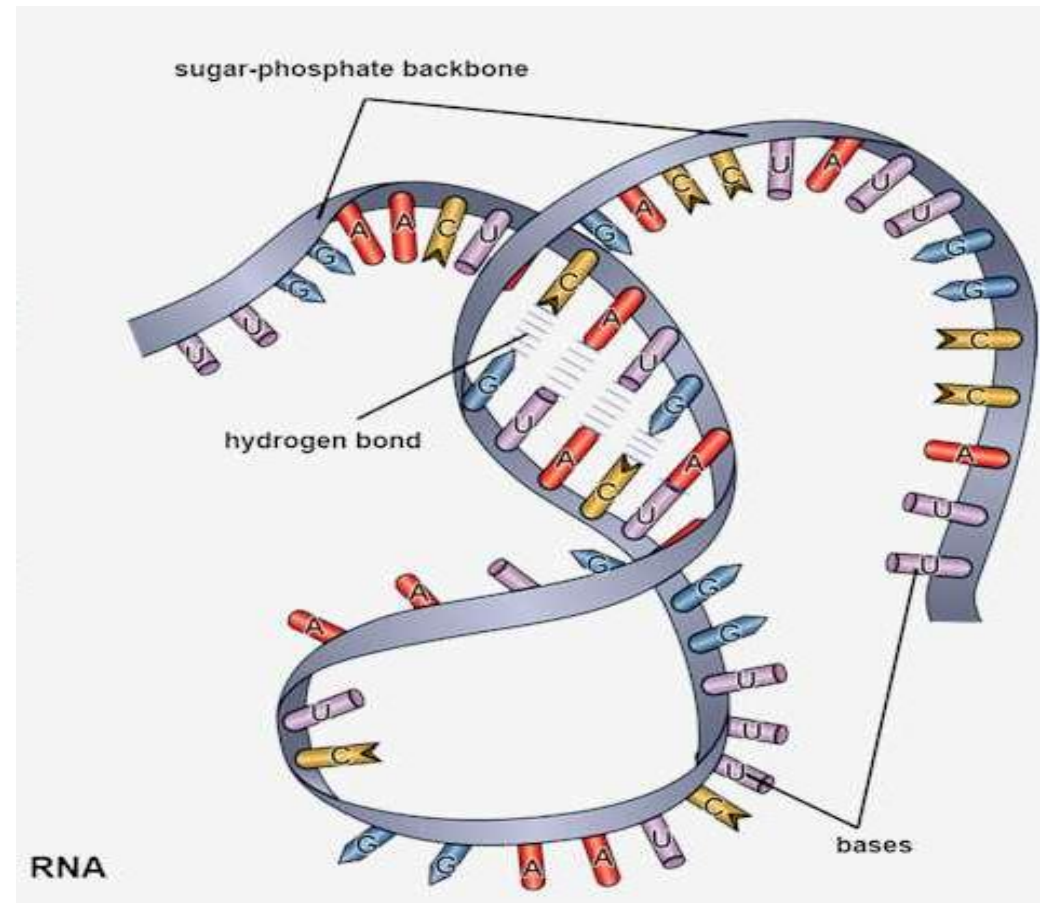
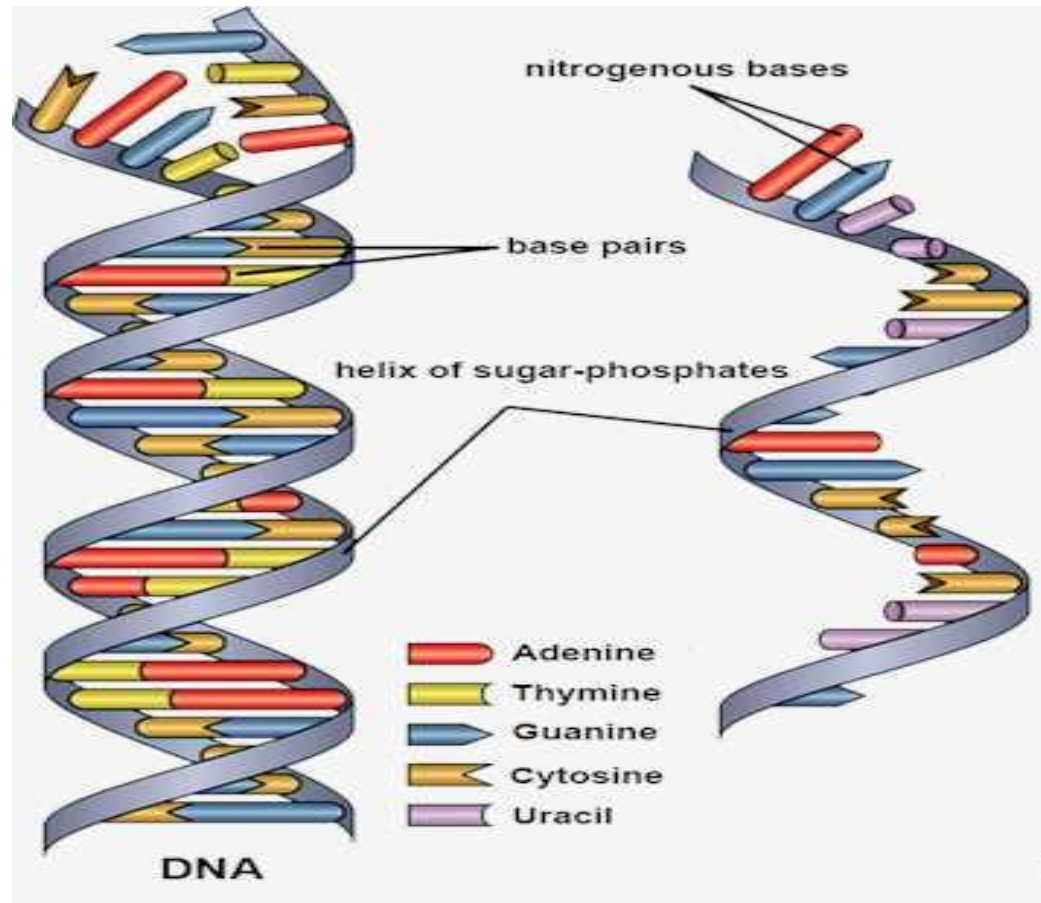
TOPICS

Types of DNA and RNA
Complementarity of DNA
Base pairing

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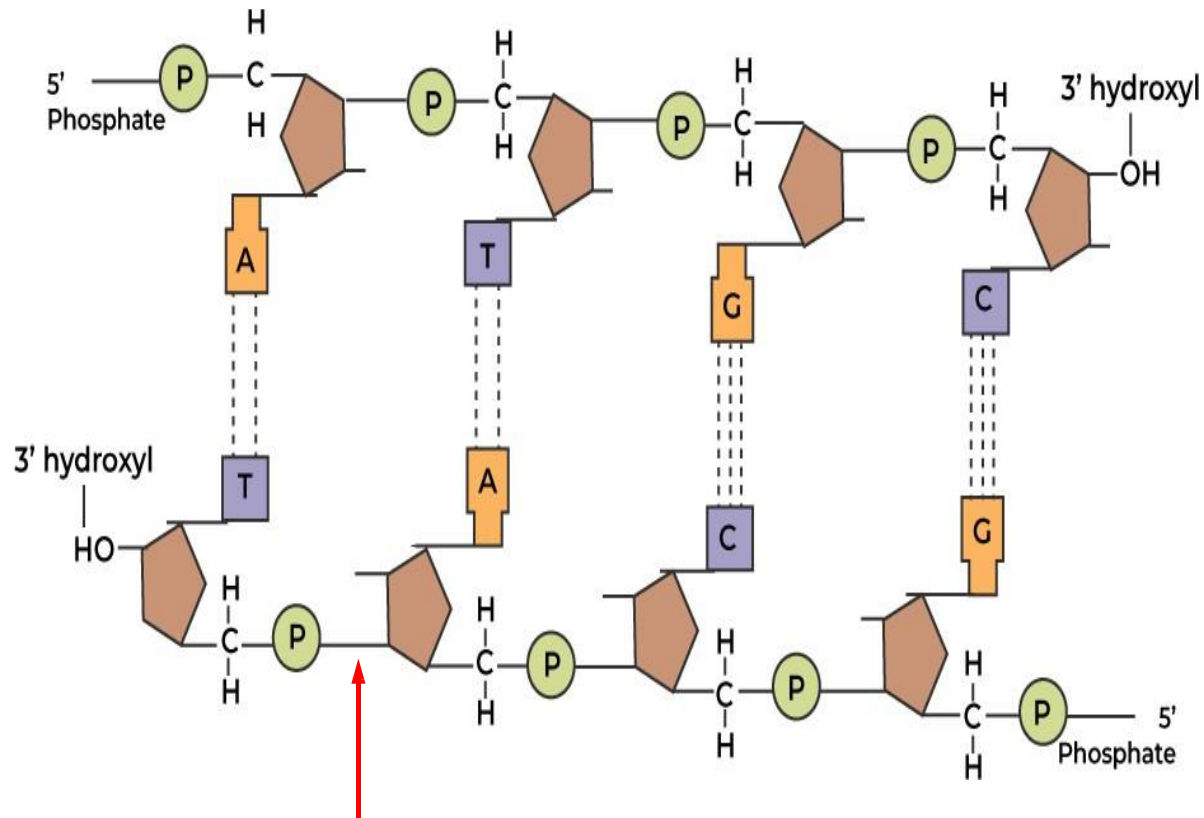
TYPES OF NUCLEIC ACIDS

(DNA & RNA)



DNA

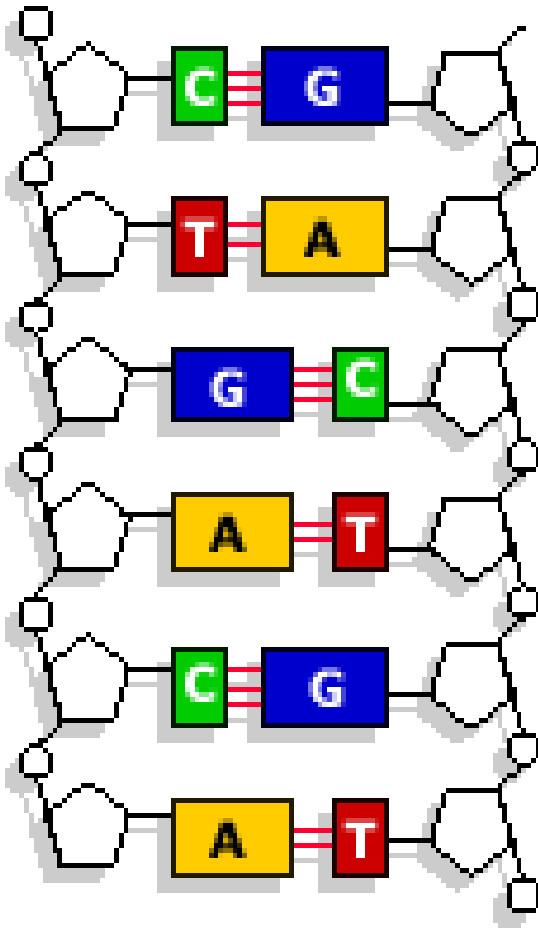
DNA (Polynucleotide Chain)



Backbone of Polynucleotide chain (Phosphate & Sugar)

3.2 DNA STRUCTURE

The two strands of a DNA are known as **polynucleotides** since they are composed of simpler units called **nucleotides**. Each nucleotide is composed of a **nitrogen-containing nucleobase**—either cytosine (C), guanine (G), adenine (A), or thymine (T)—as well as a **sugar called deoxyribose** and a **phosphate group**. The nucleotides are joined to one another in a chain by **covalent bonds** between the sugar of one nucleotide and the phosphate of the next, resulting in an alternating **sugar-phosphate backbone**. According to base pairing rules (A with T, and C with G), hydrogen bonds bind the nitrogenous bases of the two separate polynucleotide strands to make double-stranded DNA. The total amount of related DNA base pairs on Earth is estimated at 5.0×10^{37} , and weighs 50 billion tonnes. The DNA of the **largest human chromosome**, chromosome No. 1, consists of approximately **220 million base pairs** and would be 75 cm long if straightened.



Base pairs are complementary to each other. Therefore, if the sequence of one strand is known. The other strand sequence can be predicted. Because of this, the structure of DNA became clear.

DNA as an acidic substance present in nucleus was first identified by F. Miescher in 1869. However, due to **limitation in isolation** of such a long polymer intact, structure of DNA remain elusive for a long time.

It was only in 1953, James Watson and Francis crick based on the X-ray diffraction data produce by Maurice Wilkins & Rosalind Franklin, proposed the famous double helix model of structure of DNA.

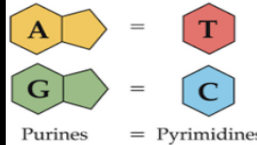
Proportion of the base pairing between 2 strands of polynucleotide chains was based on observation of Erwin Chargaff.
(Ratio between A & T, G & C are constant and equal)

Chargaff's rules

Erwin Chargaff proposed two rules which are termed as *Chargaff's rules*. These rules played an important role in the discovery of the double helix structure of DNA.

CHARGAFF'S RULE SUMMARY

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Rule 1: The amount of Adenine ~equals the amount of Thymine
The amount of Guanine ~equals the amount of Cytosine
The amount of purine = the amount of pyrimidine

Rule 2: The amount of A+T $\neq$ amount of G+C.
This ratio varies among different organisms,
but same in different tissues of the same organism

Erwin Chargaff (1950) determined that
in any strand of DNA:

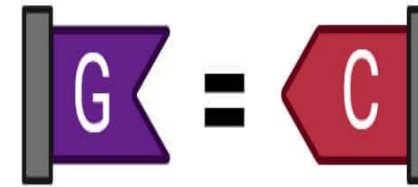
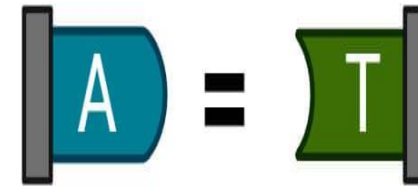
$$\% \text{ purines} = \% \text{ pyrimidines}$$

$$\% \text{A} = \% \text{T}$$

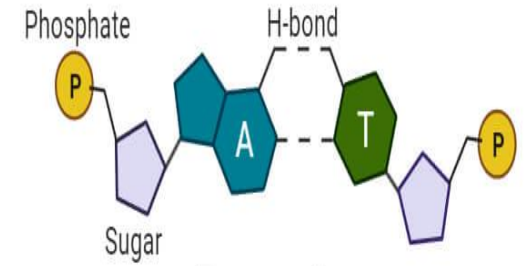
$$\% \text{C} = \% \text{G}$$

$$\# \text{A} + \text{T} \neq \# \text{C} + \text{G}$$

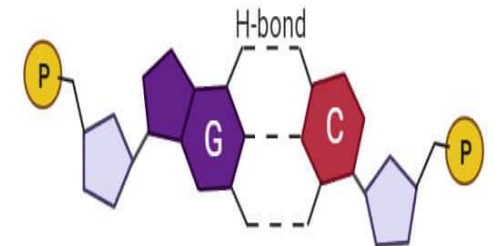
Chargaff's Rule



Purines = Pyrimidines



Base pair
(A-T interactions)



Base pair
(G-C interactions)

Chargaff's rule

- ① Amount of (A) = Amount of (T)
- ② Amount of (C) = Amount of (G)
- ③ Amount of Purine = Amount of Pyrimidine

$$A + G = C + T$$

$$\Rightarrow \frac{A+G}{C+T} = 1$$

$$④ A + T + C + G = 100$$

(Ratio between A & T, G & C are constant and equal)

- Q. In ds DNA has 40% thymine. Calculate the percentage of Guanine.
- 30%
- 40%
- 20%
- 10%

Q. DNA has 15% Adenine. Find out the amount of other bases?

Ans: A = 15%

Acc. to Chargaff's rule A = T
 $\Rightarrow T = 15\%$

$$A + T + C + G = 100$$

$$15 + 15 + C + G = 100$$

$$C + G = 100 - 30 = 70$$

A DNA molecule has 180 base pairs and 20% adenine.

How many cytosine nucleotides are present in this molecule of DNA?

$$20\% A - T \quad 20\%$$

$$30\% G - C \quad 30\%$$

$$180 \times 2 = 360$$

$$360 \times 0.3 = 108$$

BASE PAIRING

Base pairing is the specific hydrogen bonding between **nitrogenous bases** in the two strands of DNA that holds the **double helix** together.

DNA's structure, as proposed by **Watson and Crick (1953)**, is a **double-stranded helical** molecule.

Each strand is made of repeating nucleotides:

- **Sugar** → deoxyribose
- **Phosphate group** → forms the backbone
- **Nitrogenous base** → adenine (A), thymine (T), guanine (G), or cytosine (C)

The bases of one strand pair with the bases of the opposite strand in a specific manner.

A = T pair

Adenine's amino group ($-NH_2$) hydrogen bonds to thymine's carbonyl oxygen ($=O$)

Thymine's nitrogen atom hydrogen bonds to adenine's nitrogen

Two Hydrogen Bonds form between A-T base pair.

G ≡ C pair

Guanine's amino group hydrogen bonds to cytosine's carbonyl oxygen

Cytosine's amino group hydrogen bonds to guanine's carbonyl oxygen

A third hydrogen bond forms between G-C base pair.

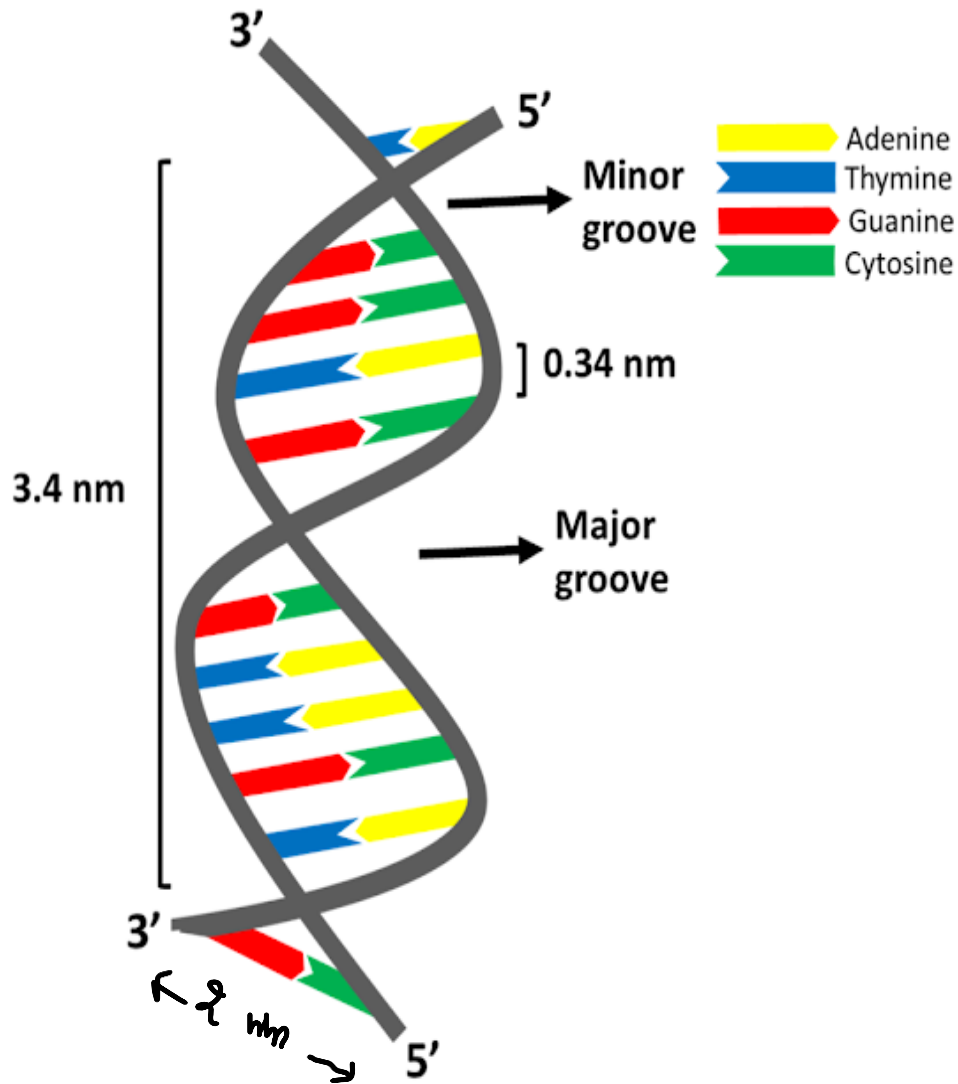
More hydrogen bonds = stronger stability

G-C pairs contribute more to DNA's melting temperature (T_m) than A-T pairs.

The two DNA strands run in opposite directions ($5' \rightarrow 3'$ and $3' \rightarrow 5'$)

Base pairing only works properly when the strands are **antiparallel**,
because the hydrogen bonds can align correctly.

Double helical structure of DNA



Watson and Crick DNA

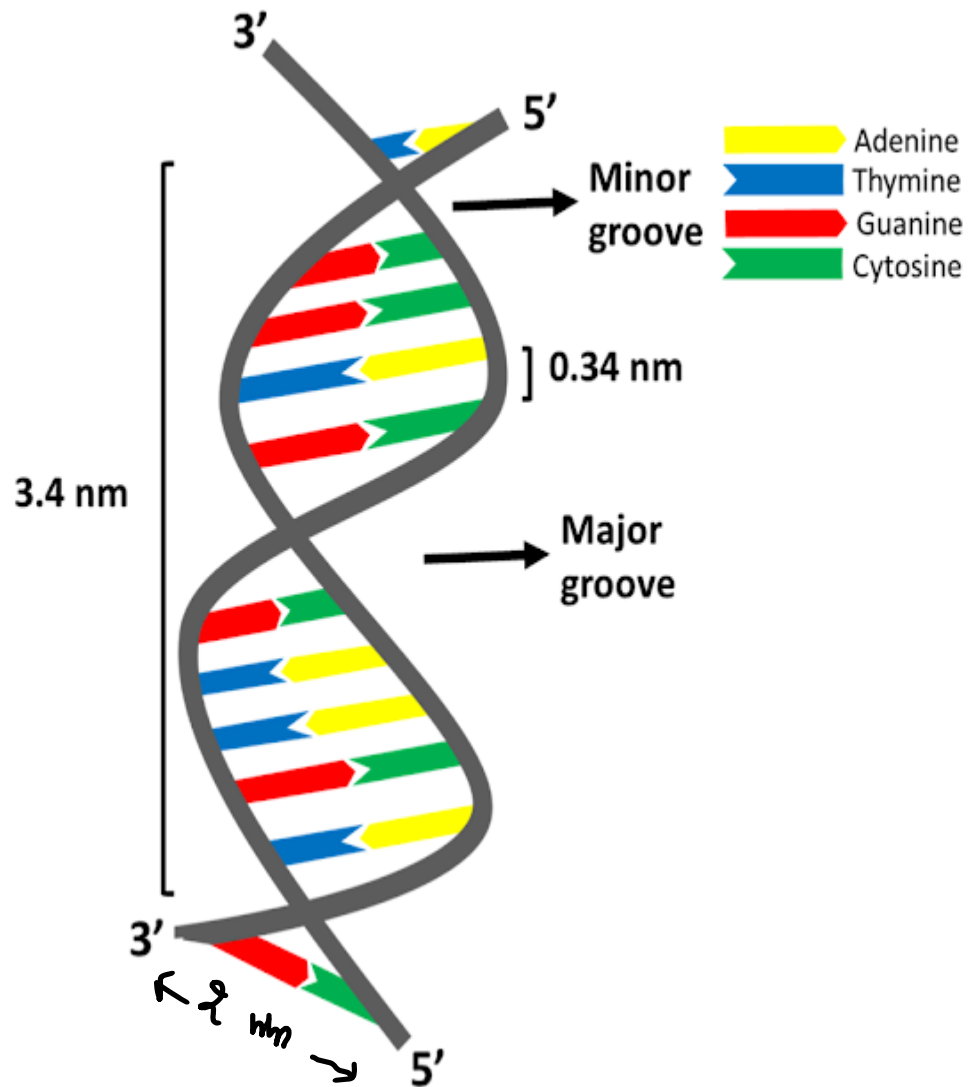
The salient features of the Double-helix structure of DNA are as follows:

1. The DNA double helix is made of two polynucleotide chains of deoxyribonucleotides.
2. Each chain of the double helix comprises of the backbone of sugar-phosphate groups and the nitrogenous bases projecting inside.
3. The two chains have anti-parallel polarity. It means, if one chain has the polarity $5' \rightarrow 3'$, the other has $3' \rightarrow 5'$.
4. The bases in two strands are paired through hydrogen bond (H-bonds) forming base pairs (bp).
 - Adenine forms two hydrogen bonds with Thymine from opposite strand and vice-versa. Similarly, Guanine is bonded with Cytosine with three H-bonds.
 - Thus, always a purine comes opposite to a pyrimidine. This generates approximately uniform distance between the two strands of the helix of 2.0 nm.
5. The two chains are coiled in a right-handed fashion.

The pitch of the helix is 3.4 nm (a nanometre is one billionth of a metre, that is 10^{-9} m) and there are roughly 10 bp in each turn. Consequently, the distance between a bp in a helix is approximately equal to 0.34 nm.
6. The plane of one base pair stacks over the other in double helix. This, in addition to H-bonds, confers stability of the helical structure.

SALIENT FEATURES OF DNA DOUBLE HELIX

Double helical structure of DNA



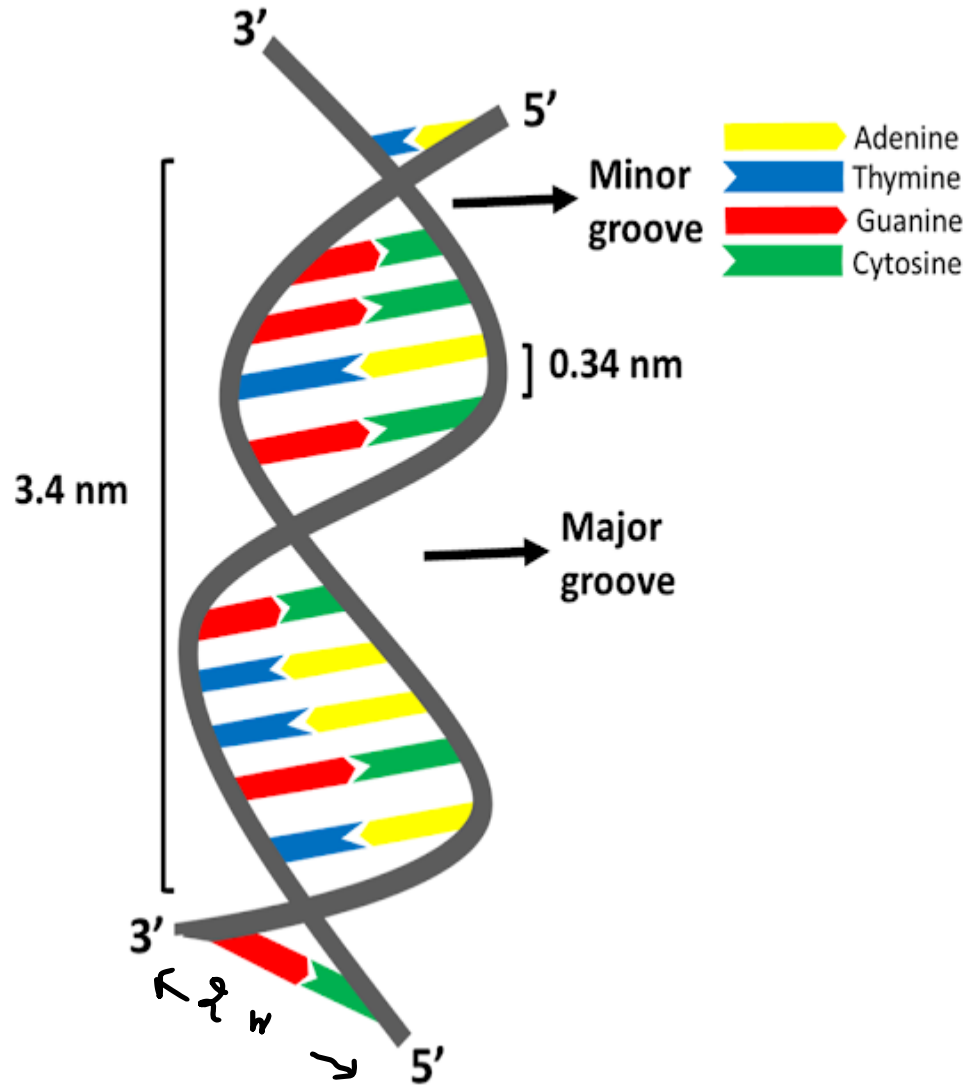
Watson and Crick DNA

SALIENT FEATURES OF DNA DOUBLE HELIX

The following are some of the important characteristic features of a DNA double helix as proposed by Watson and Crick

1. A DNA molecule is made up of two long strands of polydeoxyribonucleotides coiled in a helical way around an **imaginary central axis**.
2. The DNA duplex is coiled plectonemically in a **right handed** manner (now called **B-DNA**) just as a rope stair is twisted to form a spiral. It has a diameter of 20 Å (thus radius 10 Å).
3. The helical coiling produces alternate **major** (length 22 Å) and **minor** (length 12 Å) **grooves**.
4. Each turn of the helix has a distance of 34 Å (3.4 nm) which contains 10 nucleotides in each chain so that the average distance between adjacent nucleotide is 3.4 Å (0.34 nm).
5. Four types of deoxyribonucleotides occur in DNA (i) **dAMP** (deoxyadenosine monophosphate or adenylic acid), (ii) **dGMP** (deoxyguanosine monophosphate or guanylic acid), (iii) **dCMP** (deoxycytidine monophosphate or cytidylic acid) and (iv) **dTMP** (deoxythymidine monophosphate or thymidylic acid).
6. Each deoxyribonucleotide has three components: (i) deoxyribose sugar ($C_5H_{10}O_4$), (ii) nitrogenous base (4 types) and (iii) phosphoric acid (H_3PO_4). Nitrogenous bases may be 9-membered double ring purine which may be adenine (A) or guanine (G) and 6-membered single ring pyrimidine which may be cytosine (C) and thymine (T).
7. Deoxyribose sugar and phosphoric acid form the backbone of DNA strands while nitrogenous base lies at right angle to it, being attached at 1' carbon of the sugar, through a glycosidic bond ($-C-N-$) either at N-1 in case of a pyrimidine (T or C) or N-9 in case of a purine (A or G).
8. **Phosphodiester linkages** or bonds (2 ester formation by the same phosphate radical) are formed between 5'C of sugar of one nucleotide and 3'C of sugar of another nucleotide (hence known as 3', 5'-phosphodiester bond).

Double helical structure of DNA



10. Complementary Base Pairing. The nitrogenous bases of the two opposite strands pair in a complementary way *i.e.*, a purine pairs with a pyrimidine and *vice-versa*. Hence adenine of one chain pairs by two hydrogen bonds with thymine of the other chain (one H—O and one N—H hydrogen bond). Similarly, guanine pairs with cytosine by three hydrogen bonds (two H—O and one N—H). Thus, the two strands are called complementary to each other.

11. Two types of forces stabilize the DNA helix and hold the two chains together *i.e.* (i) hydrogen bonds between the complementary nitrogen bases and (ii) hydrophobic interactions between nitrogen bases. The nitrogen bases are stacked inside the helix while the polar groups are kept on the outside in contact with water.

12. Sense and Antisense Strand. Out of the two strands of the DNA, only one strand carries the genetic information which is called template or antisense strand. The other strand is mirror copy

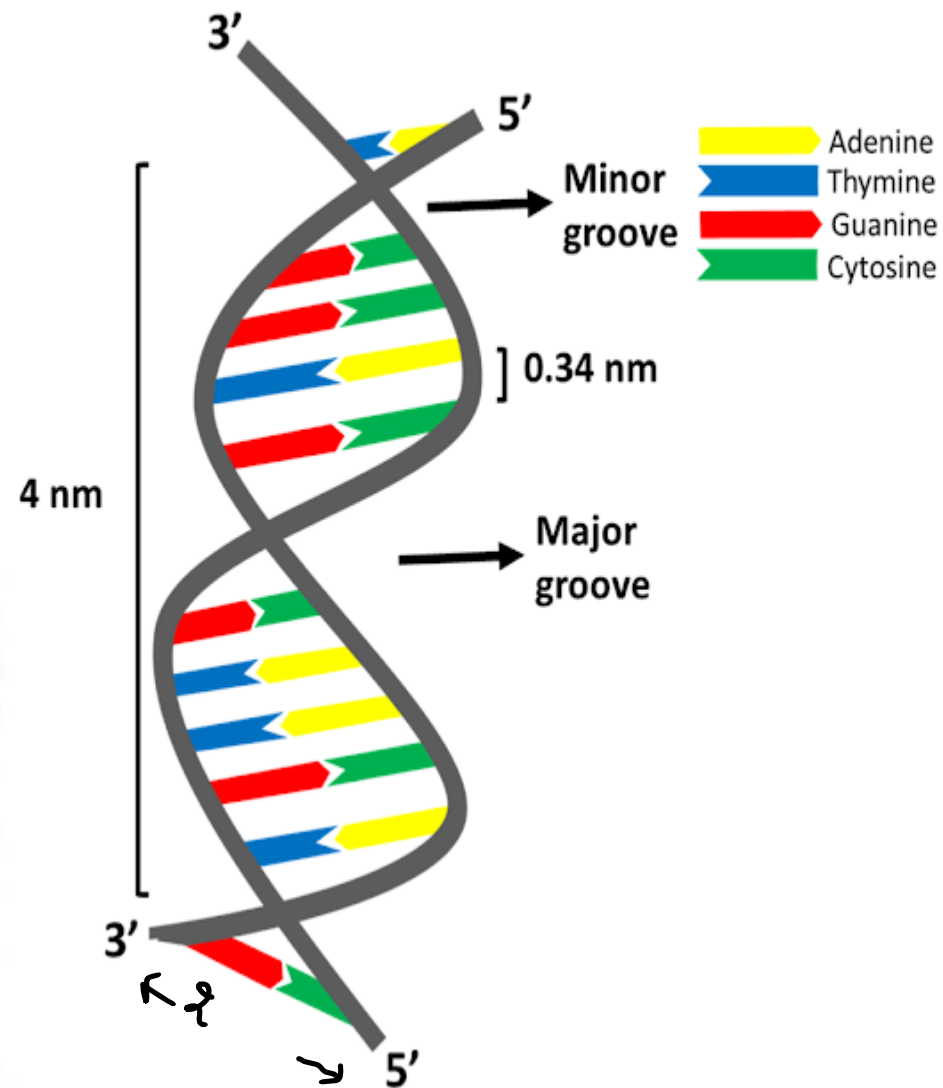
Watson and Crick DNA

which is called **coding** or **sense strand**. The sense strand covers the nitrogen bases of antisense strand and protects it from cellular biomolecules. Complementarity occurs between the antisense strand of DNA and the only one strand of mRNA formed over it *i.e.* U, A, C, G are formed on mRNA opposite to A, T, G, C of DNA antisense strand respectively. Thus, the sequence of **sense strand of DNA is same as that of mRNA**.

13. The Watson-Crick model of DNA double helix is right handed *i.e.* it runs in clockwise direction looking along the helical axis. This model is a '**B**' form of DNA which is thought to prevail in the living cell. This model is in its B lattice configuration found in the fibres of very high relative humidity (92%) and solutions of low ionic strength.

14. **Supercoiling of DNA:** DNA can be twisted like a rope in a process called DNA supercoiling. With DNA in its "**relaxed**" state, a strand usually circles the axis of the double helix once every 10.4 base pairs, but if the DNA is twisted the strands become more tightly or more loosely wound. If the DNA is twisted in the direction of the helix *i.e.*, clockwise turns of the right handed helix this is **positive supercoiling**, and the bases are held more tightly together. If they are twisted in the opposite direction (anti-clockwise) this is **negative supercoiling**, and the bases come apart more easily (Fig. 3.12). In nature, most DNA has slight negative supercoiling that is introduced by enzymes called **topoisomerases**. These enzymes are also needed to relieve the twisting stresses introduced into DNA strands during processes such as transcription and DNA replication. Supercoiling is also found in closed circular DNA of prokaryotes which is generally negative found *in vivo*. Positive supercoiling can be created *in vitro*.

Double helical structure of DNA



Watson and Crick DNA

TYPES OR DIFFERENT FORMS OF DNA

(A, B, C, D, E and Z-DNA)

DNA does not exist only in the **B-form** proposed by Watson and Crick. DNA may exist in some other forms depending on the **no. of nucleotides per turn** and the **distance between repeating units or nitrogenous bases**.

Depending on:

- **Hydration level**
- **Salt concentration**
- **DNA sequence**
- **Direction of supercoiling**
- **Chemical modifications of the bases**
- **and the type and concentration of the metal ions in the solution**

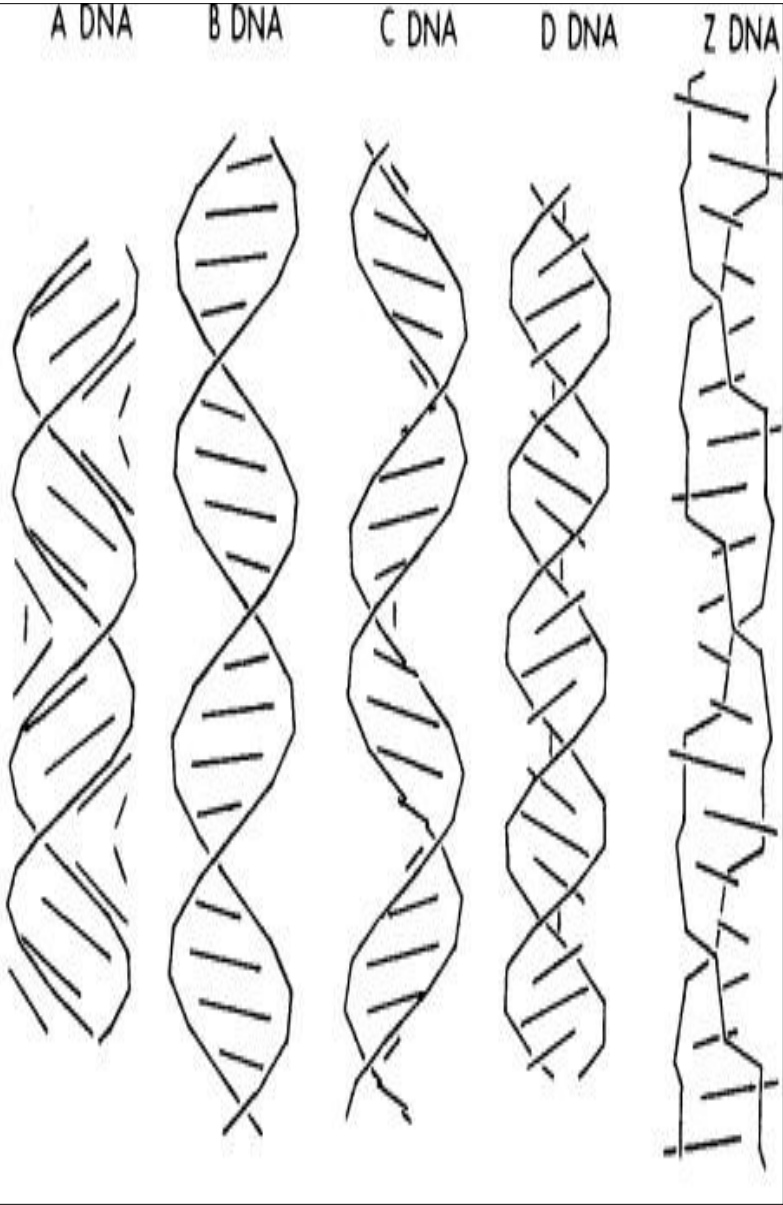
DNA can adopt different conformations.



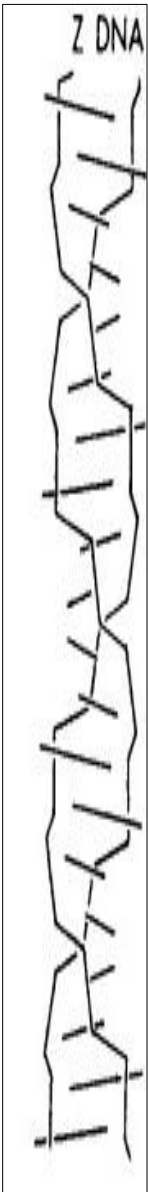
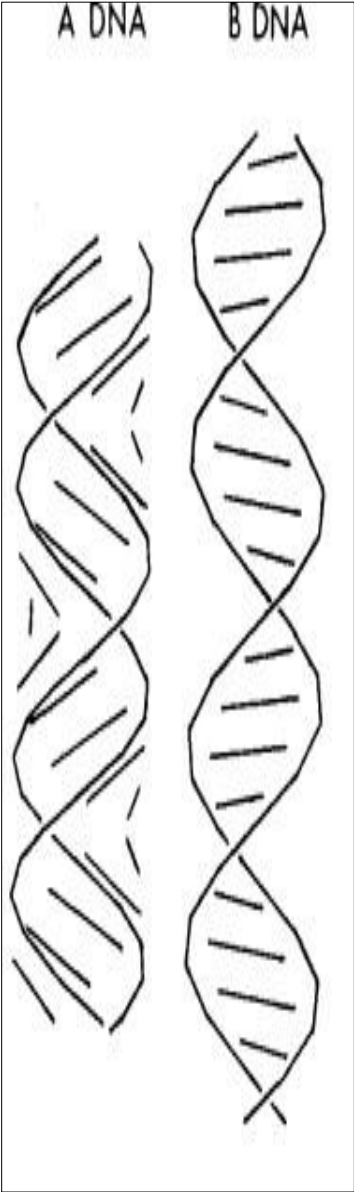
<u>DIFFERENT FORMS OF DNA</u>	
Right-handed	Left-handed
A-DNA	Z-DNA
B-DNA	(Zig-zag)
C-DNA	
D-DNA	
E-DNA	

Comparison between different forms of DNA

Parameters	A-DNA	B-DNA	C-DNA	D-DNA	E-DNA	Z-DNA
Conditions for different forms of DNA To study their structure	75% relative humidity Na ⁺ , K ⁺ , Cs ⁺ may be present as counter ions	High hydration (≈92% RH), low ionic strength	When DNA fibres are maintained in 66% RH with Li ⁺ ions C DNA is obtained	Relative humidity low, DNA lacking guanine	Low DNA lacking guanine	Very high salt concentration
Occurrence or Found	Found in dehydrated DNA & DNA-RNA hybrid duplexes	Normally found in living cell. Most common in cells	Moderate dehydration	Rare under normal conditions	Very rare	Rare, GC-rich DNA
Shape	Short, broad/wide helix	intermediate shape	More compact than B-DNA	Very compact helix	Extreme compact form	Zig-zag sugar-phosphate backbone
Helix direction/coiling/turn	Right handed	Right handed	Right handed	Right handed	Right handed	Left handed
Base pairs/turn	11	10	~9.3	8	7.5	12
Diameter	~2.6 nm (26 Å°)	~2.0 nm (20 Å°)	~1.9 nm (19Å°)	Smaller	Smaller	~1.8 nm (18 Å°)
Major groove	Narrow, deep	Wide, deep	Narrow	Narrow	Narrow	Flat (Z DNA possess only one type of grove
Minor groove	Wide, shallow	Narrow	Narrow	Narrow	Narrow	Single groove corresponds to minor groove

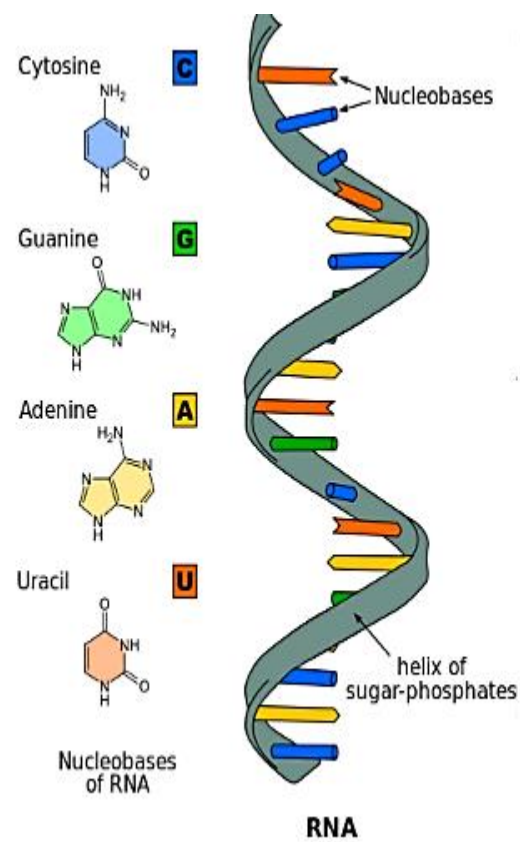


DNA can exist in multiple conformations. But most important conformations are **A, B and Z DNA**



Sl No	Parameters	A-DNA	B-DNA (Physiological form)	Z-DNA
1	Helix coiling	Right-handed	Right-handed	Left-handed
2	Occurrence	Low humidity or hydration (Dehydrated) DNA-RNA hybrids	Normal cellular condition Predominant (Watson & crick DNA)	- High salt concentration - GC rich regions favors Z-DNA formation
3	Shape	Short & Broadest	Intermediate	Narrowest
4	sugar phosphate backbone	Regular	Regular	Zigzag
5	Repeating units	Mononucleotide	Mononucleotide	Dinucleotide
6	Base pairs/turn	11	10	12
7	Pitch/ height of the helix (A°)	28 A° or 2.8nm (Shortest)	34 A° or 3.4nm (Intermediate)	45 A° or 4.5 nm (Longest)
8	Distance between 2 base pairs (A°)	2.5 A°	3.4 A°	3.7 A°
9	Rotation per base pair	+32°	+36°	-30°
10	Diameter of helix A°	~2.6 nm (26 A°)	~2.0 nm (20 A°)	18 A°
11	Grooves (Major & Minor)	Major: Narrow very deep Minor: very broad shallow	Major: wide deep Minor: narrow deep	Major: flat Minor: Narrow deep
12	Sugar pucker	C3'- endo	C2' endo	Alternating (C3'- endo & C2' endo)

RNA

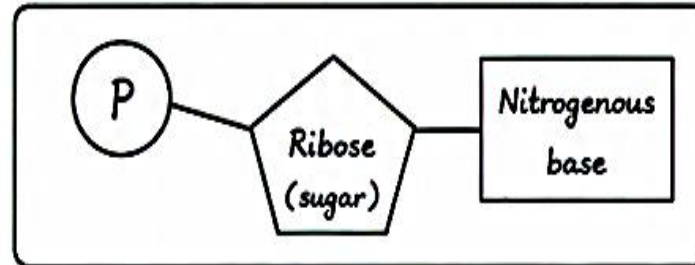


≡ THE RNA ≡

Definition :

RNA (Ribonucleic Acid) is a nucleic acid present in almost all living cells. It carries genetic information from DNA and is involved in protein synthesis.

Structure of RNA :



- Made up of ribonucleotides.
- Each nucleotide has:
 - A phosphate group
 - Ribose sugar (with 2'-OH group)
 - A nitrogenous base (A, U, G, or C)
- RNA is single stranded but can form secondary structures (hairpin loops, etc.).

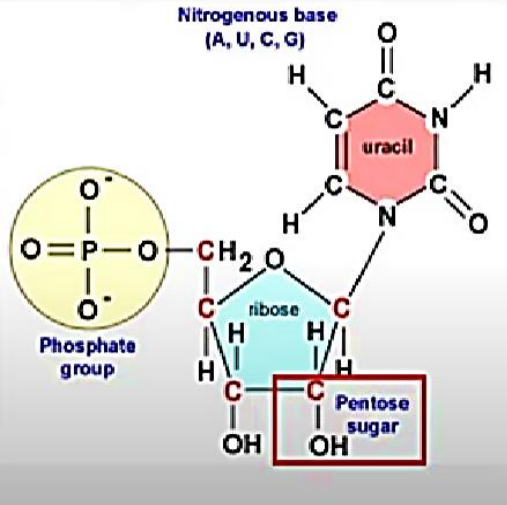
Key Points :

- Discovered by Severo Ochoa in 1955.
- Composed of ribonucleotides.
- Usually single stranded.
- Sugar → Ribose
- Bases → Adenine (A), Uracil (U), Guanine (G), Cytosine (C)

RNA (Ribose Nucleic Acid) is a single strand polyribonucleotide present in the cytoplasm and nucleus of all living organisms. In the cytoplasm it may be associated with ribosome. Very rarely it is double stranded as in case of **Reovirus** in animals and **Wound Tumor Virus** in plants. Each ribonucleotide has 3 components i.e. (i) Ribose ($C_5H_{10}O_5$) sugar (ii) Nitrogenous bases (adenine, guanine, cytosine and uracil) and (iii) phosphoric acid. These structures are similar to DNA except that instead of deoxyribose sugar, ribose sugar and instead of thymine, uracil is present in RNA. Ribonucleotides are also joined by 3', 5'-phosphodiester bonds like that of DNA.

Note :

RNA is less stable than DNA because of the 2'-OH group in ribose. It is more reactive and easily degraded by RNases.



TYPES OF RNA

(Ribonucleic Acid)

Depending upon the hereditary information, RNA may be of two types as follows:

1. Genetic or Genomic RNA: The RNA which acts as genetic material is called genetic RNA present in riboviruses and viroids. In viroids, RNA has both genetic and catalytic function. Genetic RNA is single stranded in most cases *i.e.*, Tobacco Mosaic Virus (TMV), influenza virus, Human Immunodeficiency Virus (HIV) etc. It is double stranded in Reovirus, Wound Tumour Virus and Rice Dwarf Virus etc.

2. Non-genetic RNA: These RNA molecules do not act as genetic material, rather they are associated with protein synthesis. They are synthesized from DNA by the process of transcription.

2. Non-genetic RNA: These RNA molecules do not act as genetic material, rather they are associated with protein synthesis. They are synthesized from DNA by the process of transcription.

Types of RNA

In all prokaryotic and eukaryotic organisms, three main classes of RNA molecules exist-

- 1) Messenger RNA(m RNA)
- 2) Transfer RNA (t RNA)
- 3) Ribosomal RNA (r RNA)

The other are –

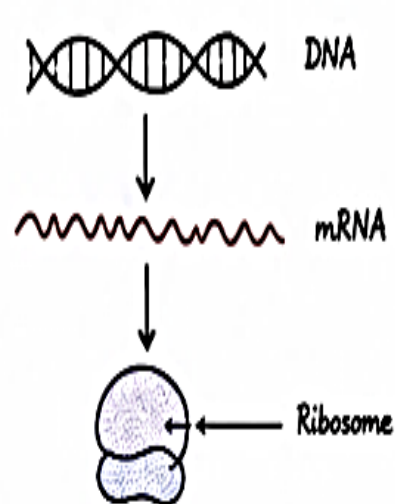
- small nuclear RNA (SnRNA),
- micro RNA(mi RNA) and
- small interfering RNA(Si RNA) and
- heterogeneous nuclear RNA (hnRNA).

RNA	Size and length	Percent of total cellular RNA	Function
rRNA	28 S, 18 S, 5.8 S, 5 S (23 S, 16 S, 5 S)*	80	Interact to form ribosomes
tRNA	65–110 nt	15	Adapter
mRNA	0.5–6+ kb	5	Directs synthesis of cellular proteins

Types of RNA :

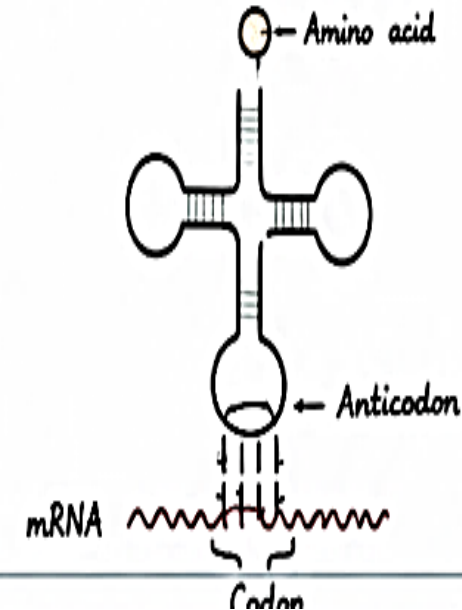
1. mRNA (Messenger RNA)

- Carries genetic information from DNA to ribosomes.
- Acts as a template for protein synthesis.



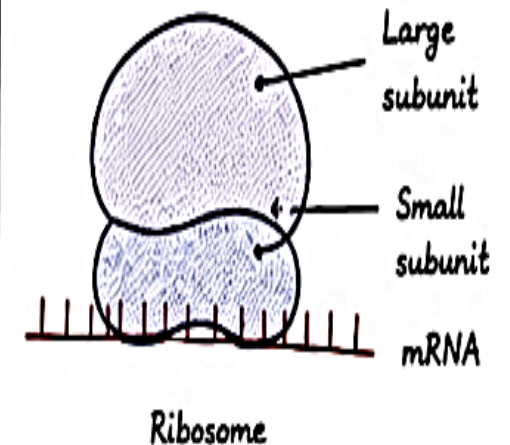
2. tRNA (Transfer RNA)

- Carries specific amino acids to the ribosomes.
- Has an anticodon that pairs with codon on mRNA.



3. rRNA (Ribosomal RNA)

- Major component of ribosomes.
- Provides structural support and helps in protein synthesis.

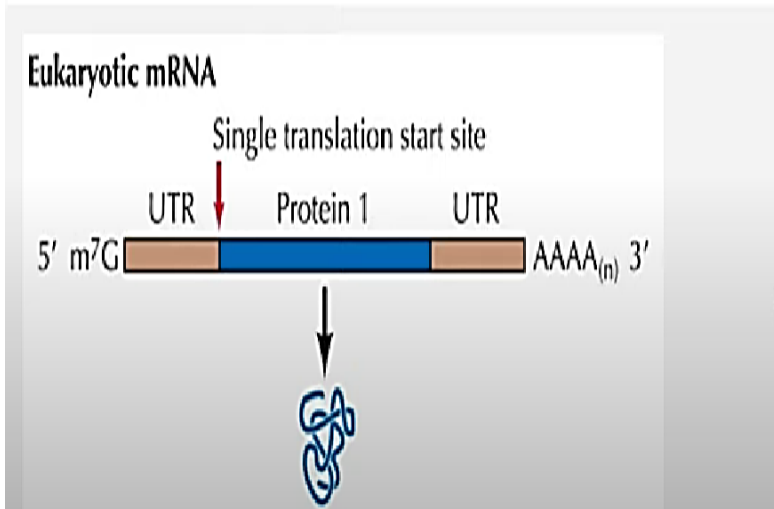
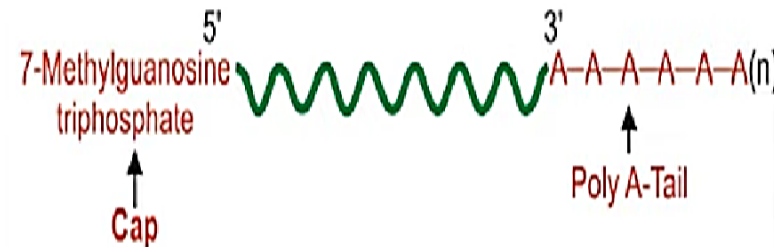


Type Of Non-Genetic RNA (Ribonucleic Acid)

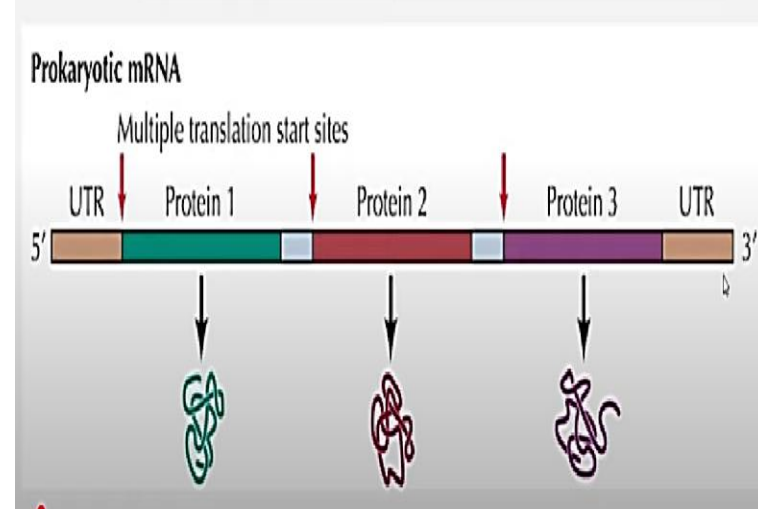
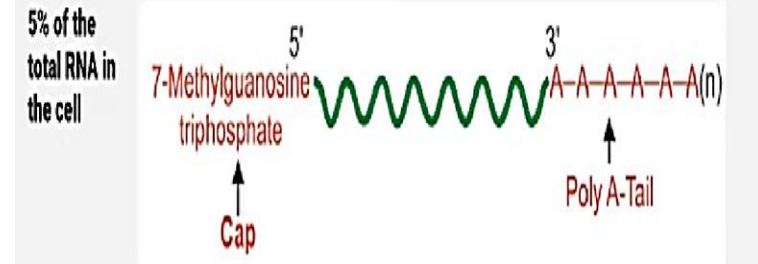
- mRNA (Messenger RNA), Informational RNA or Template RNA.
- It brings coded information from DNA for protein synthesis.
- It is a long RNA having molecular weight from 5,00,000 to 20,00,000 and constitutes only 5% of the total RNAs.

- The mRNA may be monocistronic carrying information for one polypeptide or polycistronic carrying information for many polypeptides.
- mRNA is more rapidly produced in the cell but it exists for a much shorter period of time than rRNA and tRNA.
- Since each mRNA is short lived, the total amount of mRNA present at any time is very low.
- The mRNA molecule is dismantled immediately after it has been used for synthesis of a particular protein for which it is short-lived.

1) mRNA (MESSENGER RNA)



MONOCISTRONIC m-RNA



POLYCISTRONIC m-RNA

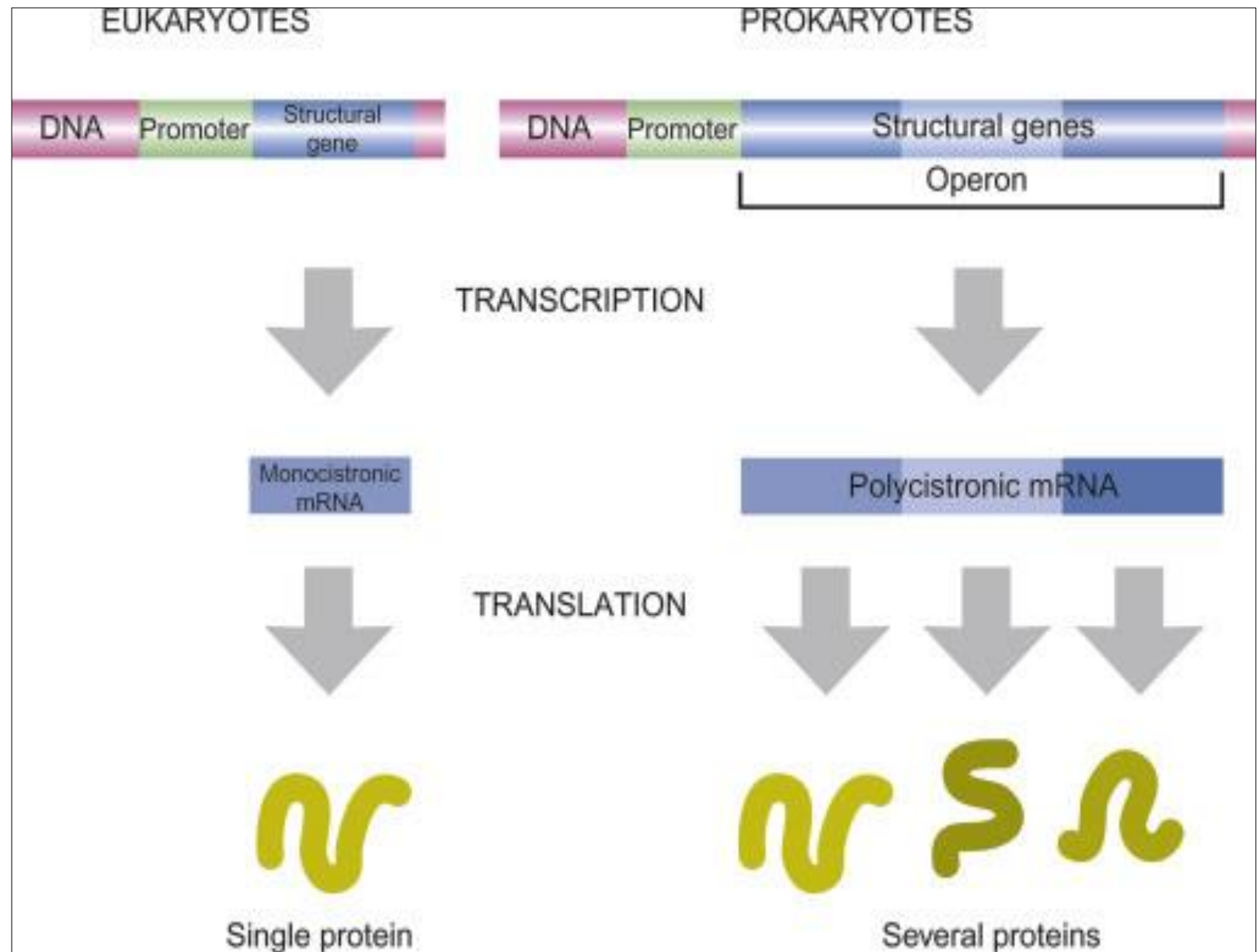
Cistron: an old name for a gene

Polycistronic: 1 promoter directs synthesis of 1 mRNA that can be translated to more than one polypeptide

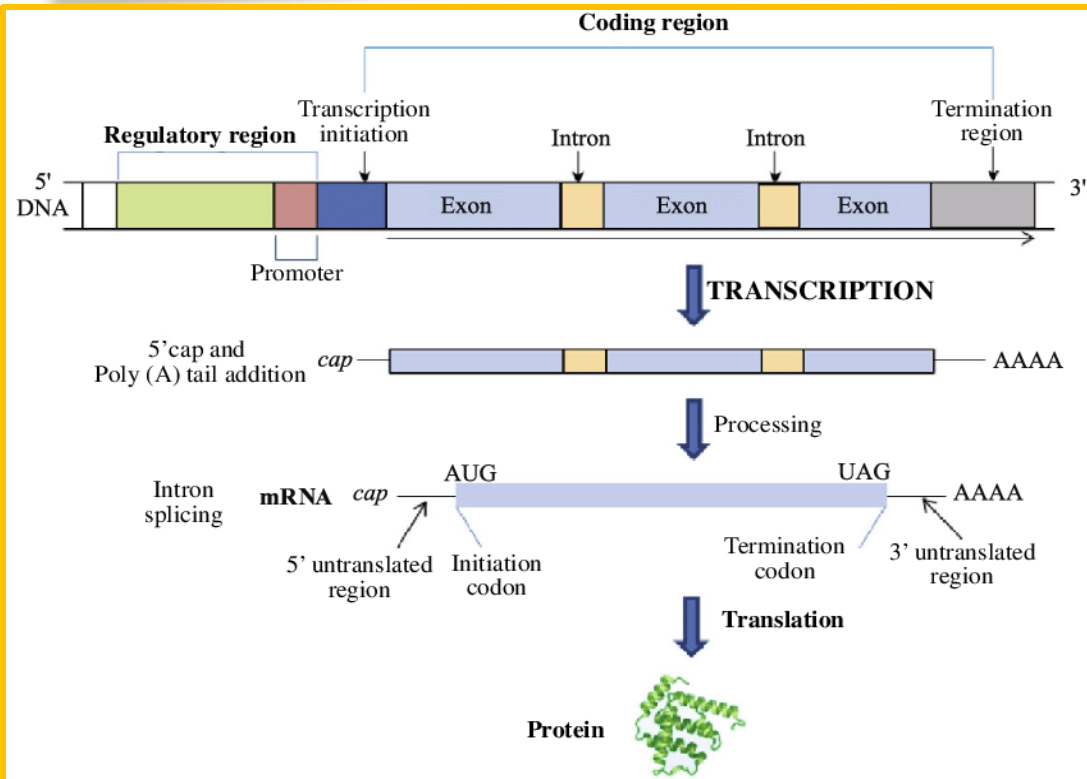
- Prokaryotic genes

Monocistronic: 1 promoter directs synthesis of 1 mRNA that **usually** translates to only 1 protein

- Eukaryotic genes



mRNA (MESSENGER RNA)



Structure & Features of mRNA :

- It is single stranded non-genetic RNA.
- It contains a **cap region** with modified GTP at the 5' end for attaching to ribosome and initiating translation.
- Also another cap is present at the 3' end with 20-25 adenine nucleotides (the **poly-A tail**).
- The mRNA also contains **initiation codon** (AUG or GUG), **coding region** having numerous triplet codons and in the end **termination codon** (UAA, UAG or UGA).
- It contains numerous triplet codons.
- Short lived and rapidly degraded after use.

A schematic representation of Eukaryotic mRNA



- Recognition of mRNA in protein biosynthesis
- Prevent from 5'-exonucleases activities

- Prevent from 3'-exonucleases activities
- Helps in exporting mRNA from nucleus to cytosol

2. Ribosomal RNA (rRNA)

- rRNA is a type of non coding RNA which is a part of the ribosome.
- rRNA is formed in the nucleolus region of eukaryotes and cytoplasm in prokaryotes
- Its molecular weight varies from **35,000 to 10,00,000**.

rRNAs combine with proteins in the cytoplasm to form ribosomes, which act as the site of protein synthesis and has the enzymes needed for the process.

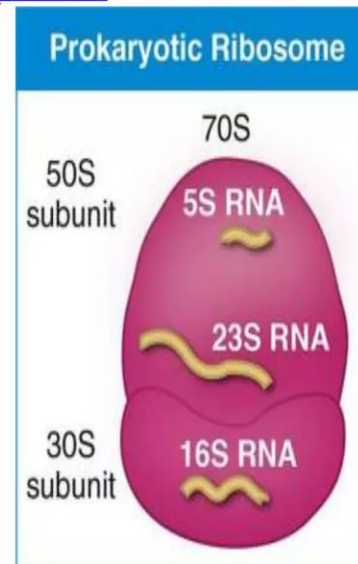
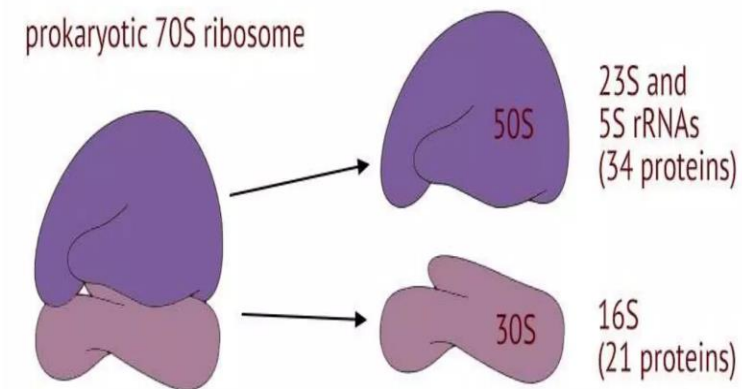
➤ Ribosomes are composed of approximately 60% rRNA and 40% ribosomal proteins by mass.

(rRNA is involved in **protein synthesis**)

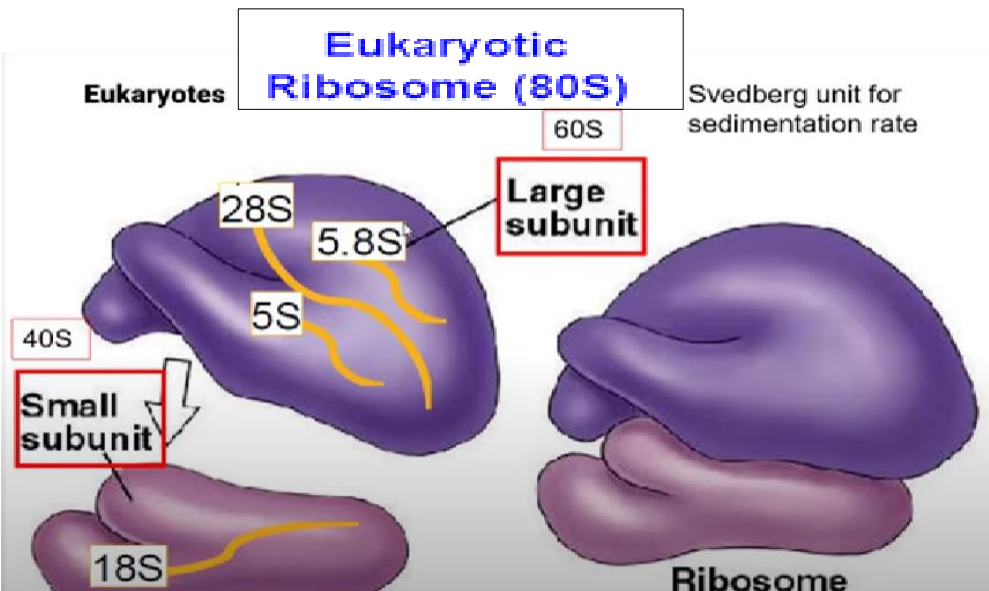
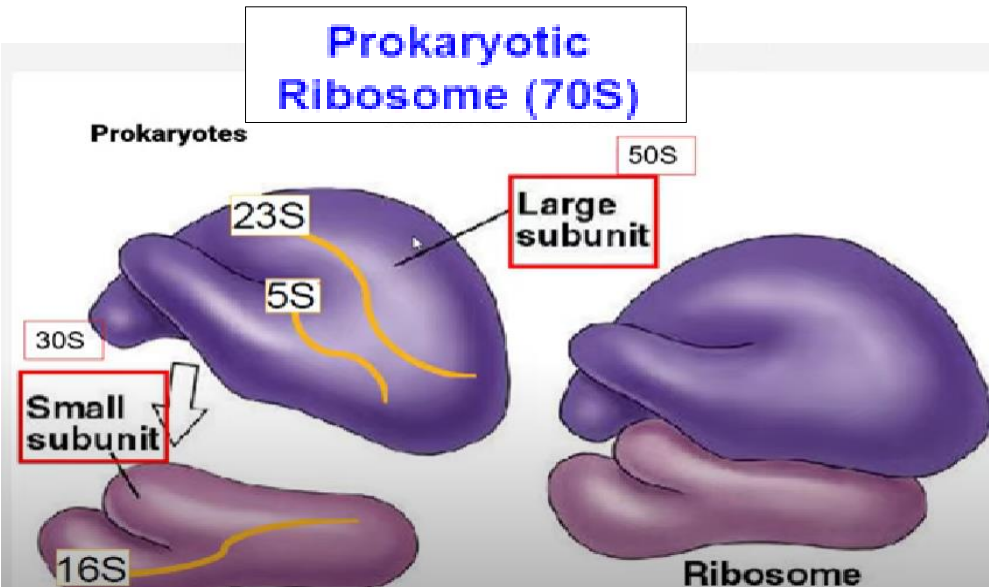
- rRNA is a ribozyme which carries out protein synthesis in ribosomes.
- Ribosomal RNA is the predominant form of RNA found in most cells; it makes up about 80% of cellular RNA despite never being translated into proteins itself.

Types of rRNA

- Ribosomal RNA organizes into two ribosomal subunits: the large ribosomal subunit ([LSU](#)) and small ribosomal subunit ([SSU](#)). Between these subunits, the rRNA types used to form the subunit differ.
- In the ribosomes of prokaryotes such as [bacteria](#), the SSU contains a single small rRNA molecule (~1500 nucleotides) while the LSU contains one single small rRNA and a single large rRNA molecule (~3000 nucleotides). These are combined with ribosomal [proteins](#) to form ribosomal subunits.



Ribosome has 2 units



2. Ribosomal RNA (rRNA)

Both prokaryotic and eukaryotic ribosomes can be broken down into two subunits, one large and one small.

Type	Size	Large subunit (LSU rRNA)	Small subunit (SSU rRNA)
Prokaryotic	70S	50S (5S : 120 nt, 23S : 2906 nt)	30S (16S : 1542 nt)
Eukaryotic	80S	60S (5S : 121 nt, 5.8S : 156 nt, 28S : 5070 nt)	40S (18S : 1869 nt)

❑ All of the ribosomal RNA molecules except the 5S rRNA are processed from a single 45S precursor RNA molecule in the nucleolus .

❑ 5S rRNA is independently transcribed.

Different types of rRNAs have **specific functions**.

- **18S rRNA** provides a binding site to **mRNA**.
- **5S rRNA** has a similar binding site for **tRNA**.

● The 3' end of the 16S ribosomal RNA (in a ribosome) recognizes a sequence on the 5' end of mRNA called the Shine-Dalgarno sequence.

- rRNA is **more stable than mRNA**.
- In some cases, rRNA may have a **hairpin-like structure**.

3. Transfer RNA (tRNA)

Introduction

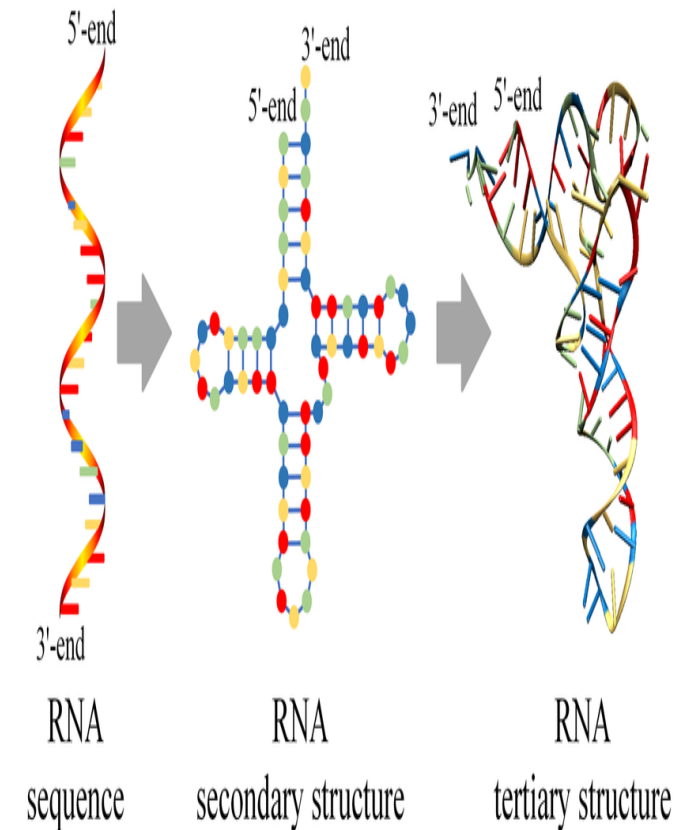
1. Transfer RNA (tRNA) is **the smallest** of the three major species of RNA molecules. **tRNAs contain 74–95 nucleotide residues.**
2. tRNA was first discovered by **Hoagland et al. in 1957.**
3. The molecular weight of tRNA is 23,000 to 30,000 daltons (Da), **or 23–30 kDa (kilodaltons).**
4. **tRNAs form about 15% of the total cell RNA.**
5. They are synthesized by the **nuclear processing of a precursor molecule.**
6. **They transfer amino acids from the cytoplasm to the protein-synthesizing machinery, hence the name tRNA.**
7. They are easily soluble; **hence, they are called “soluble RNA” or “sRNA.”**
8. **They are also called adapter molecules** because they act as adapters for translating the sequence of nucleotides of mRNA into specific amino acids.

10. There are at **least 20 species of tRNA**, with one corresponding to each of the **20 amino acids** required for protein synthesis.

11. **The tRNA molecule is folded over itself, forming a cloverleaf-like structure in two dimensions.**

12. In three dimensions, **tRNA forms an L-shaped structure.**

13. **The cloverleaf structure was described by Holley et al. (1964), while the three-dimensional L-shaped structure was described by Klug (1974).**



Structural characteristics of t- RNA

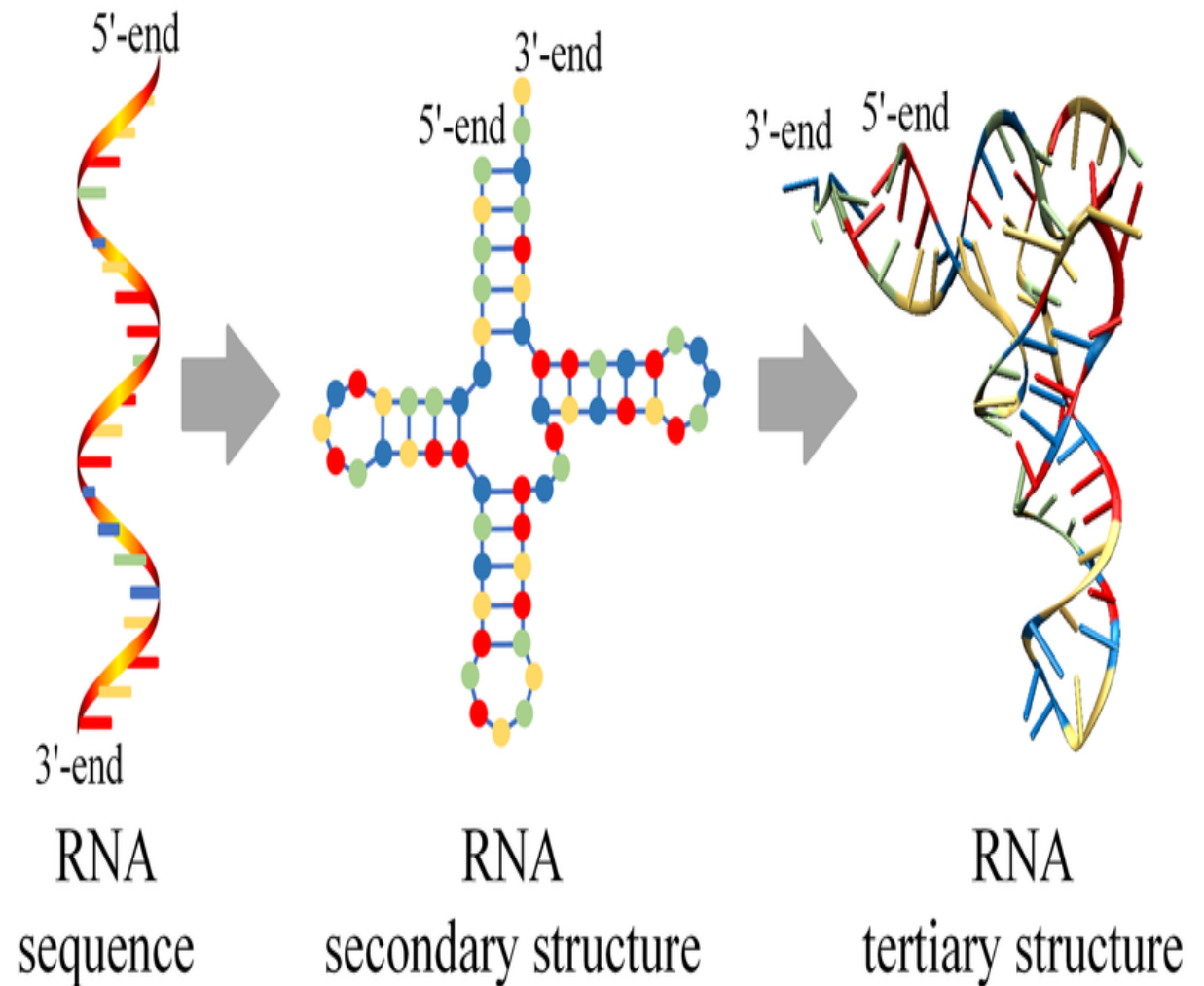
1) Primary structure- The nucleotide sequence of all the t RNA molecules allows extensive intrastand complementarity that generates a secondary structure.

2) Secondary structure- Each single t- RNA shows extensive internal base pairing and acquires a clover leaf like structure. The structure is stabilized by hydrogen bonding between the bases and is a consistent feature.

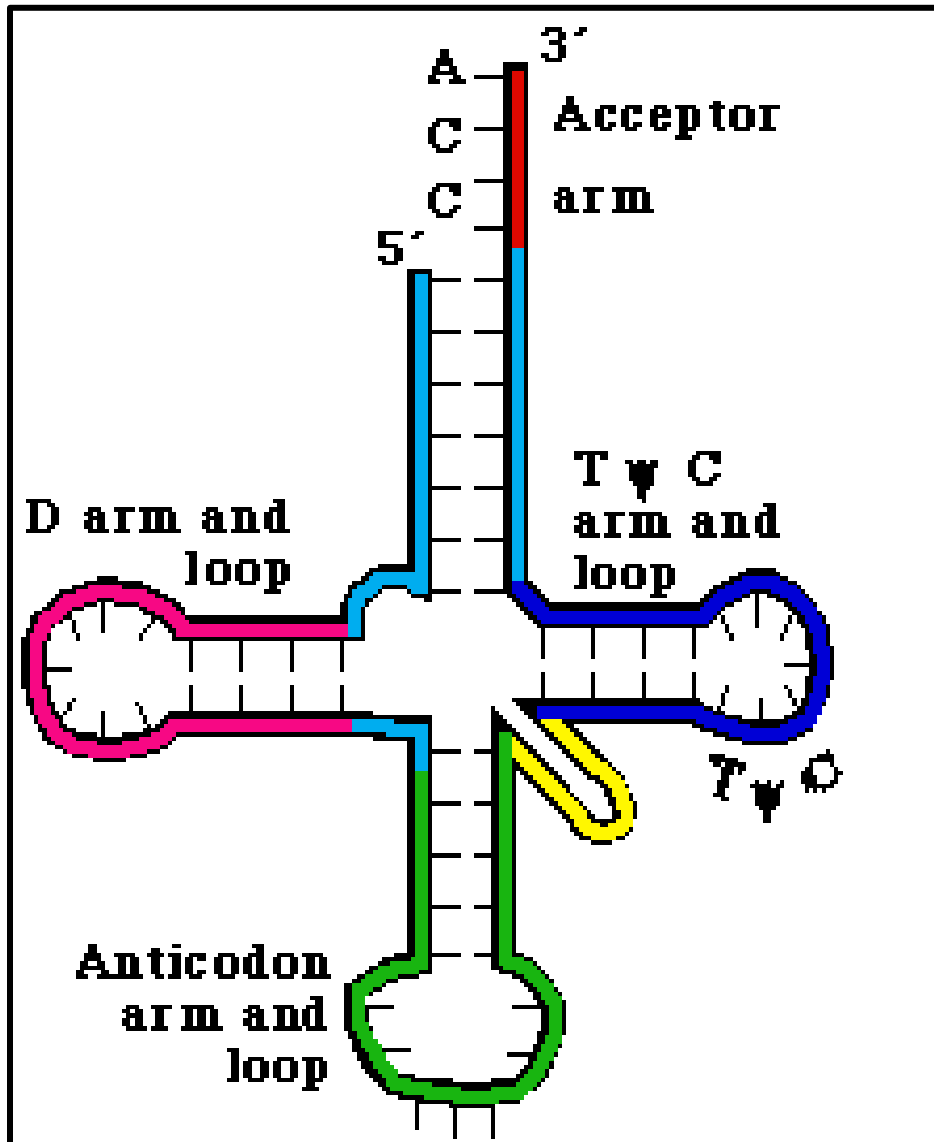
☐ The L shaped tertiary structure is formed by further folding of the clover leaf due to hydrogen bonds between T and D arms.

☐ The base paired double helical stems get arranged in to two double helical columns, continuous and perpendicular to one another.

3) Tertiary Structure



Secondary structure (Clover leaf structure)



Clover leaf structure of t-RNA

It is the secondary structure of t-RNA, that looks like a clover leaf. Hence it is named as clover leaf model of t RNA

All t-RNA contain 5 main arms or loops which are as follows-

- a) Acceptor arm
- b) Anticodon arm
- c) D HU arm
- d) TΨC arm
- e) Extra arm

A clover leaf model of tRNA has the following characteristic features:

1. It has four main arms or sites excluding a fifth small arm called extra arm or blind lump.
2. The four arms or sites are:

a) Acceptor arm

- ☐ The acceptor arm is at 3' end
- ☐ It has 7 base pairs
- ☐ The end sequence is unpaired Cytosine-Adenine at the 3' end
- ☐ The 3' OH group terminal of Adenine binds with carboxyl group of amino acid
- ☐ The tRNA bound with amino acid is called Amino acyl tRNA
- ☐ CCA attachment is done post transcriptionally

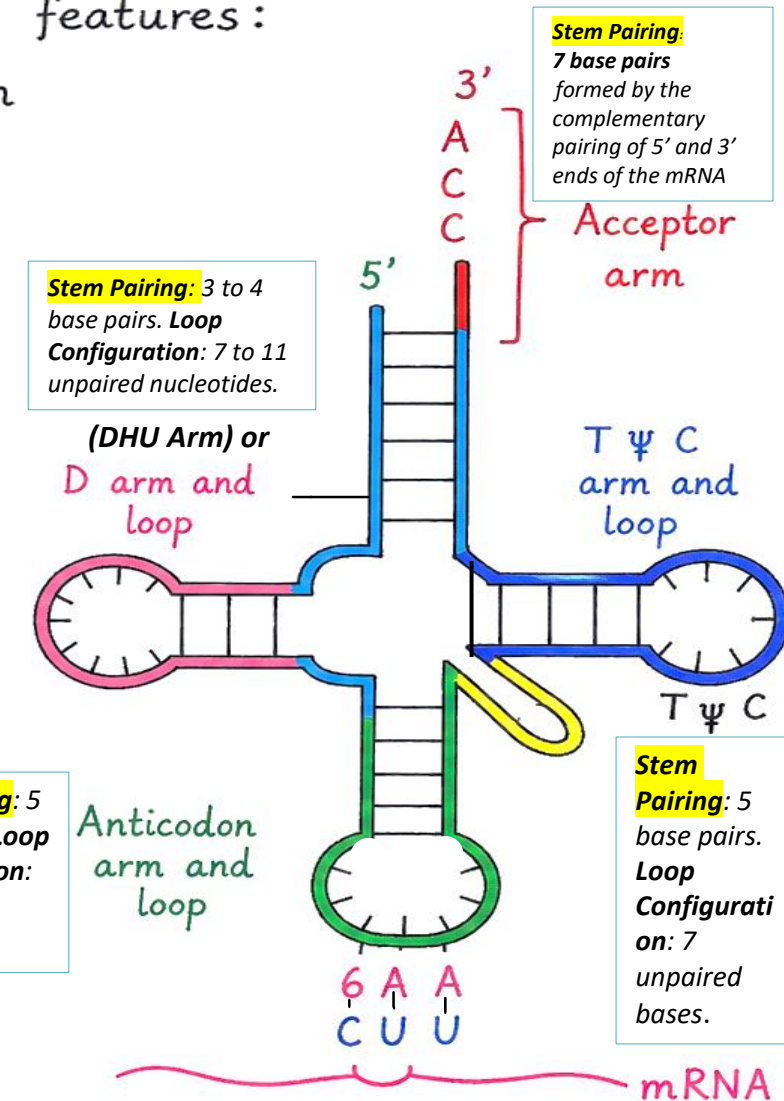
Acceptor arm or amino acid binding site having CCA - OH 3' end and G-P 5' end. The specific amino acid is bound at 3' end.

Function: The terminal Adenine residue at 3' end binds directly to the specific amino acid

b) Anticodon arm

- ☐ Lies at the opposite end of acceptor arm
- ☐ 5 base pairs long
- ☐ Base sequence of anticodon arm is complementary to the base sequence of mRNA codon.
- ☐ Due to complementarity it can bind specifically with mRNA by hydrogen bonds (For protein synthesis)

Function: The middle three nucleotides in this loop form the anticodon triplet. These bases directly recognize and pair with corresponding mRNA codons during translation.



c) DHU arm (Aminoacyl Synthetase Binding loop)

- D arm has a stem of 3-4 base pairs and it ends in a loop called **D loop** as it generally contains **dihydrouridine**, a modified nucleotide.

Stem Pairing: 3 to 4 base pairs.
Loop Configuration: 7 to 11 unpaired nucleotides.

Function:

- Serves as the **recognition site** for the enzyme (aminoacyl tRNA synthetase) that adds the amino acid to the acceptor arm.

d) T Ψ C arm (Ribosomal Binding loop)

- This arm is opposite to DHU arm
- Since it contains pseudo uridine that is why it is so named.

Stem Pairing: 5 base pairs.
Loop Configuration: 7 unpaired bases.

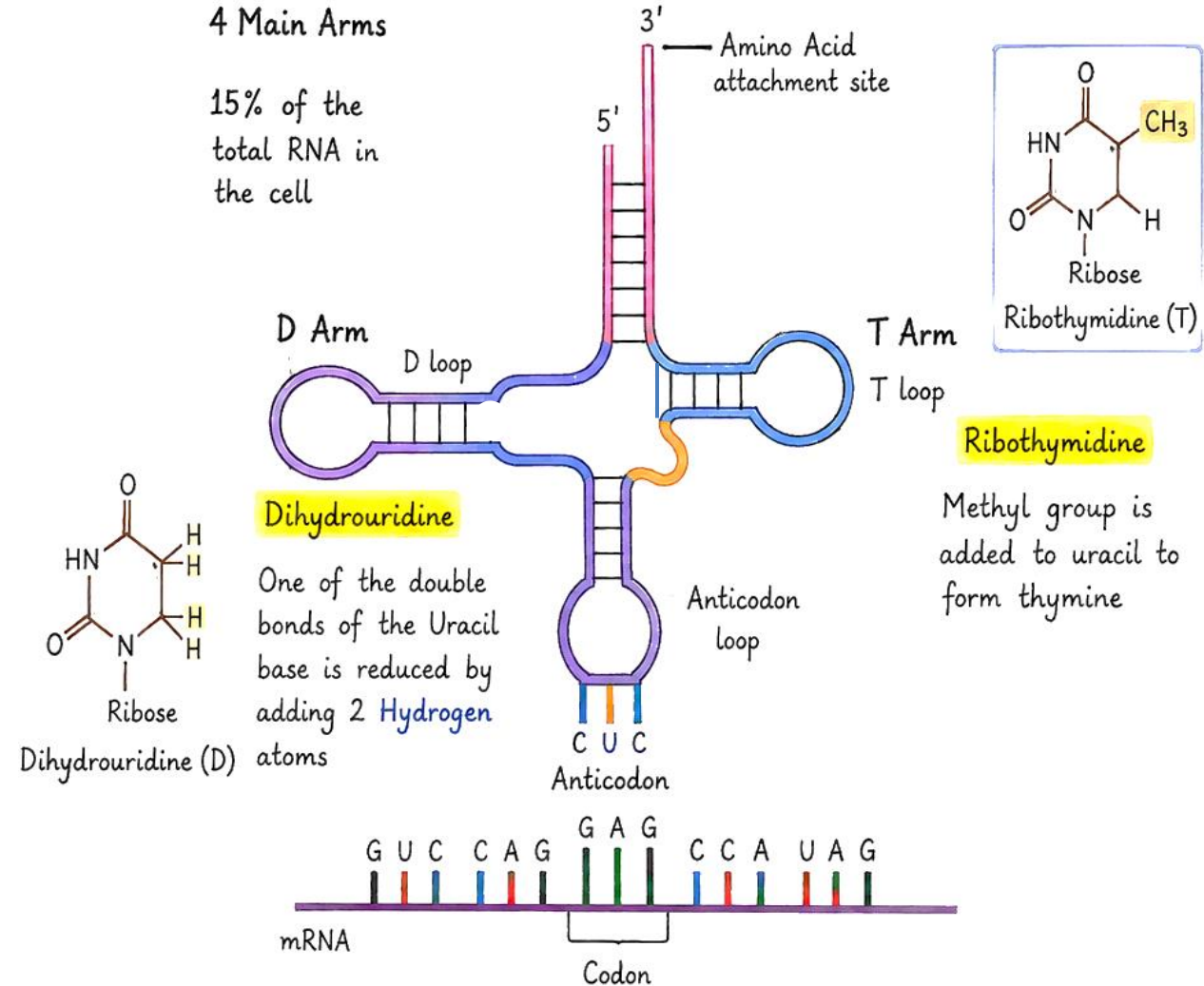
- It is involved in the binding of tRNA to the ribosomes

I.e., This loop recognizes a Ribosome which binds the tRNA in such a manner that facilitates **Codon Anticodon pairing**.

TRANSFER RNA

4 Main Arms

15% of the total RNA in the cell

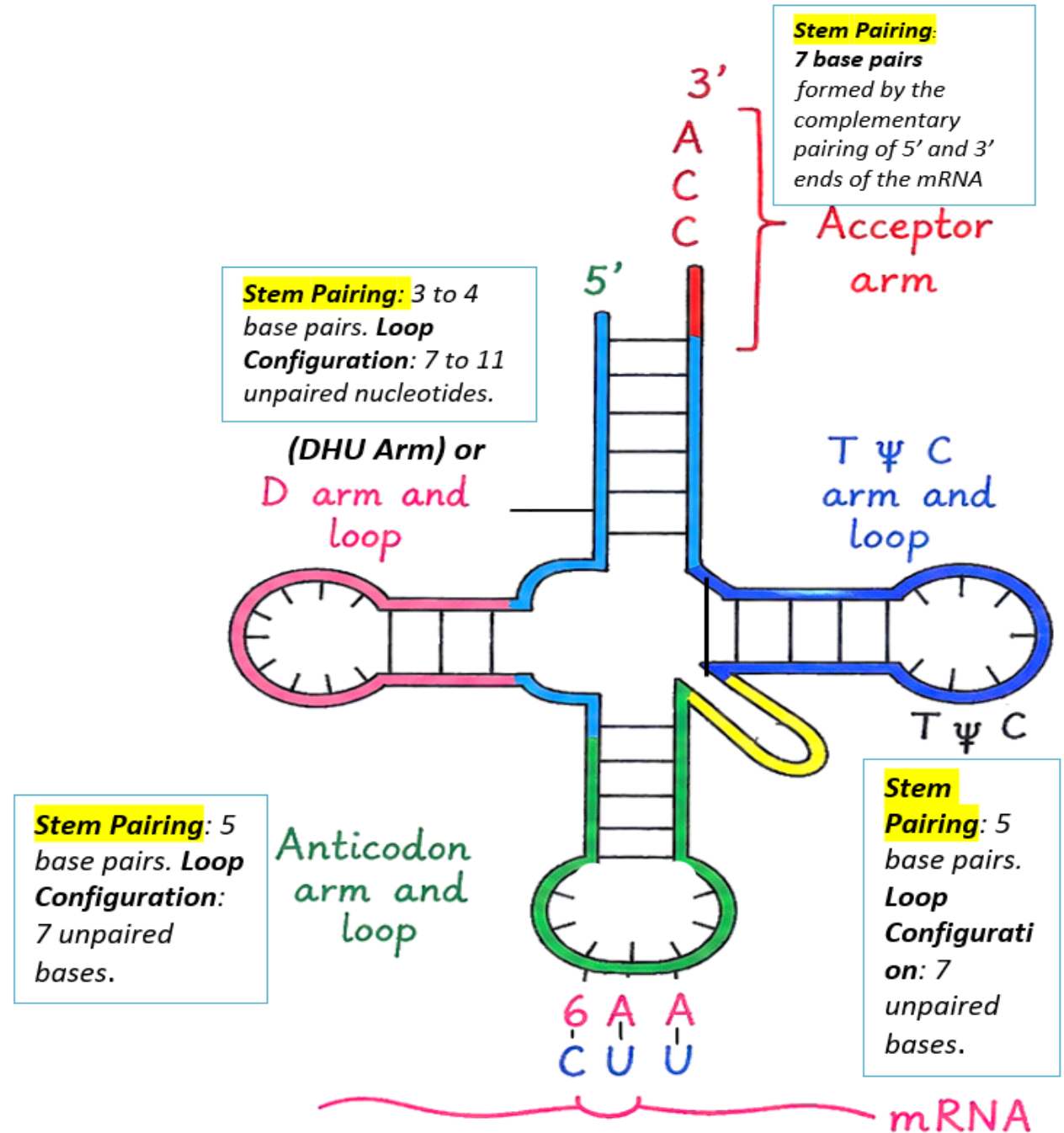


e) Extra arm or Variable arm

- About 75 % of tRNA molecules possesses a short extra arm

Stem/Loop Pairing: Highly variable configuration ranging from 3 to 21 nucleotides.

- If about 3-5 base pairs are present the t-RNA is said to be belonging to class 1. Majority t-RNA belong to class 1.
- The t-RNA belonging to class 2 have long extra arm, 13-21 base pairs in length.



OTHER TYPES OF RNA's

Types of RNA Produced in Cells

Apart from the above three types of RNAs, there are some other RNAs as follows:

1. Small Nuclear RNA (snRNA): It is a small uridine rich RNA that occurs inside the nucleus. It is associated with 7-8 molecules of proteins and takes part in splicing (formation of spliceosome) and processing of other RNAs. Examples: U1, U2, U4, U5, U6.

2. Small Cytoplasmic RNA (scRNA): It is also a small RNA present in the cytoplasm. It helps in processing of polypeptides.

3. Micro-RNA (miRNA): It is a small non-coding RNA molecule containing about 22 nucleotides and found in plants, animals and some viruses. It functions in mRNA silencing and post-transcriptional regulation of gene expression. The human genome may encode over 1000 miRNAs.

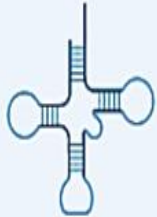
4. Small Interfering RNA (siRNA) of the RNA interference pathway. The siRNA are derived from longer regions of double-stranded RNA.

mRNA



Encodes proteins

tRNA



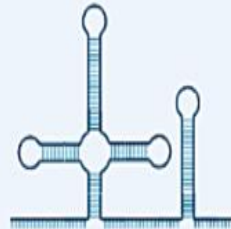
Acts as adaptor between mRNA and amino acids

rRNA



Forms the ribosome

snRNA



Functions in various nuclear processes (e.g. splicing)

miRNA



Regulates gene expression

siRNA



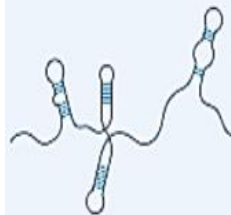
Silences gene expression

snoRNA



Facilitates chemical modification of RNAs

lncRNA



Regulates gene expression

hnRNA - heterogeneous nuclear

precursors & intermediates of mature mRNAs & other RNAs

snRNA - small nuclear

mRNA processing, poly A addition < catalytic >

snoRNA - small nucleolar

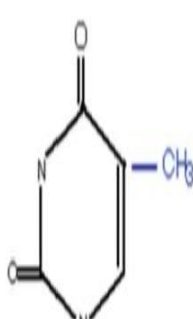
rRNA processing/maturation/methylation

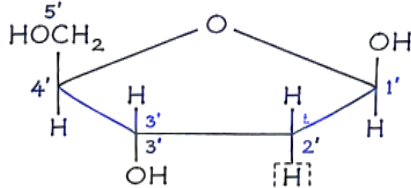
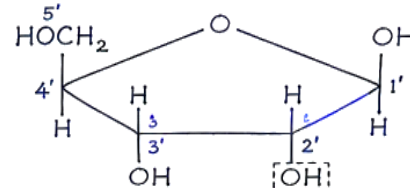
5. Guide RNA (gRNA): It is synthesized from minicircle mitochondrial DNA (mtDNA) and is used for RNA editing leading to modification of RNA transcribed from DNA template.

6. Antisense RNA (asRNA): It is a single stranded RNA that is complementary to a mRNA strand transcribed within a cell. Antisense RNA inhibits the translation of a complementary mRNA by base pairing to it and physically obstructing the translation machinery. The effects of asRNA are related with the effects of RNA interference (RNAi). It is also known as micRNA (mRNA interfering complementary RNA) as it interferes with the process of translation.

7. Short hairpin RNA (shRNA): A short or small hairpin RNA (shRNA/Hairpin Vector) is an artificial RNA molecule with a tight hairpin turn that can be used to silence target gene expression via RNA interference (RNAi).

What makes DNA and RNA different?

1) ribose sugar	RNA  (ribonucleic acid)	DNA  (deoxy ribonucleic acid)
2) T and U	 uracil	 thymine
3) strand	single	double

Sl. No.	DNA	RNA
1.	It is the genetic material of all cellular organisms.	It is the genetic material of only some viruses (riboviruses) and viroids.
2.	DNA is generally double stranded except in some viruses (colliphage ϕ X 174), where it is single stranded.	RNA is single-stranded except in some viruses (Reovirus), where it is double stranded.
3.	Mostly present in the chromosomes of nucleus, some in chloroplast and mitochondria.	Very little occur inside nucleus. major parts present in ribosomes and cytosol.
4.	The sugar present in DNA is 2-deoxyribose ($C_5H_{10}O_4$).  2-Deoxyribose	The sugar present in RNA is ribose ($C_5H_{10}O_5$).  Ribose
5.	Nitrogen bases are A, T, G, C. (Adenine, Thymine, Guanine and Cytosine).	Nitrogen bases are A, U, G, C. Here the pyrimidine uracil (U) is present instead of thymine.
6.	Number of nucleotides present in DNA is around 4 million.	Number of nucleotides present in RNA is between 70-12,000.
7.	It has ability for self-replication.	No such ability except in some viruses.
8.	Functionally DNA is of two types: nuclear and extra-nuclear.	Functionally RNA is of several types viz. genetic, messenger, transfer, ribosomal, small nuclear, small cytoplasmic and micro and small interfering RNAs etc.
9.	DNA has the ability for transcription (DNA $\rightarrow$ RNA).	It has no such ability, but can translate the coded information to form polypeptide.
10.	It exhibits positive Feulgen test (Schiffs reagent).	It shows negative Feulgen test.
11.	It does not contain unusual bases.	It contains unusual bases like pseudo-uridine, dihydrouridine, methyl-cytosine etc.
12.	Purine and pyrimidine bases are always in equal number.	There is no such fixed ratio.
13.	Nitrogen bases occur in complementary pairs in the two strands of DNA i.e., A opposite T and G opposite C.	Base pairing occurs only in certain coiled parts i.e., A opposite U and G opposite C.
14.	It is the largest macro-molecule (biopolymer) of the cell.	It is medium-sized to small sized macromolecule (biopolymer).



FUNCTIONS OF DNA

1. DNA is the genetic material of all living organisms except RNA containing viruses.
2. DNA is the **store house of genetic information** and responsible for its transmission from one generation to the other.
3. It has the capacity for **self-replication** i.e. it can reproduce its own exact copy. This characteristic feature of the DNA is the most important property of living organisms.
4. Apart from the above autocatalytic nature, DNA has **heterocatalytic function** i.e. it can produce dissimilar molecules like, RNA and proteins by transcription and translation respectively. More precisely, **transcription** involves formation of RNA from DNA, whereas **translation** involves synthesis of proteins from mRNA.
5. The **gene products are enzymes** (proteinaceous in nature), which catalyze all the biochemical reactions of the cells.
6. DNA, as it contains genes, govern all the **phenotypic characters** of an organism.
7. The process of **differentiation** in the organism is regulated by the DNA.
8. If it is damaged, it can **repair** itself so that the genetic information is preserved.
9. DNA has the possibility of **variations** by recombinations and mutations.



Functions of RNA

1. **Genomic or Genetic RNA:** These RNAs carry hereditary informations as in riboviruses and viroids.
2. **RNA as Primer:** It may act as a primer in DNA replication.
3. **In Ribosomes:** rRNAs are constituents of ribosomes.
4. **Role in Genetic Code:** mRNA contains the codons which carry genetic messages from DNA which are translated into proteins.
5. **Protein Synthesis:** All the three types of RNAs take part in the biosynthesis of proteins that are converted into enzymes or other protoplasmic structures.
6. **Polypeptide Processing:** It is carried out by small cytoplasmic RNAs (scRNAs).
7. **RNA Enzymes:** Ribozyme, a non-protein enzyme is made up of RNAs. Ribonuclease-P and peptidyltransferase enzymes are also made up of RNAs.
8. **RNA Processing:** snRNAs participate in splicing and processing of RNAs.
9. **DNA Compaction:** Small RNA strands take part in compaction of prokaryotic DNA.
10. **Informosome:** mRNA alongwith protein give rise to informosome.
11. **Micro-RNAs** functions in mRNA silencing and regulation of gene expression.