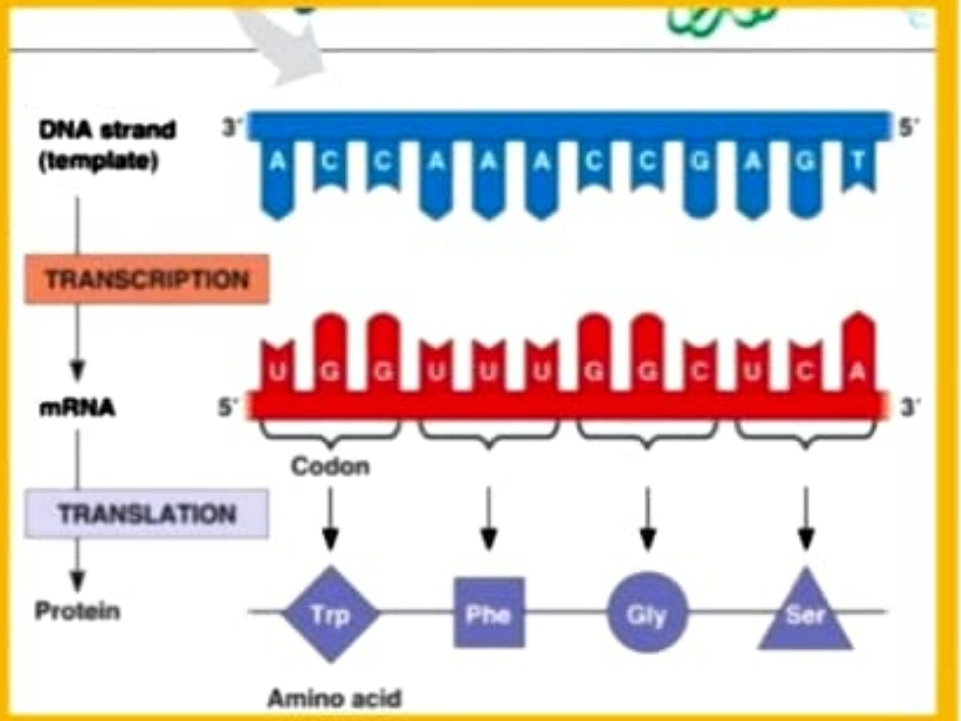
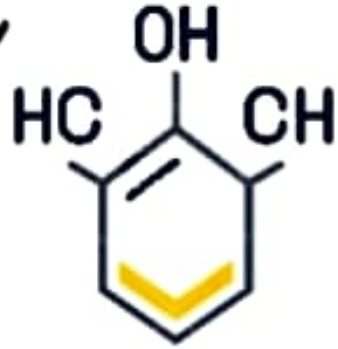
BIOLOGY



KRISHNAGAR ACADEMY



Molecular Basis Of Inheritance

Chapter - 6

CLASS - 12

The characters of a species are transmitted without much change from one generation to another. The inheritance pattern of characters indicates that their expression is controlled by genes which are located on chromosomes and constitute total genomic make-up of the organism *(Fig. 1)*.

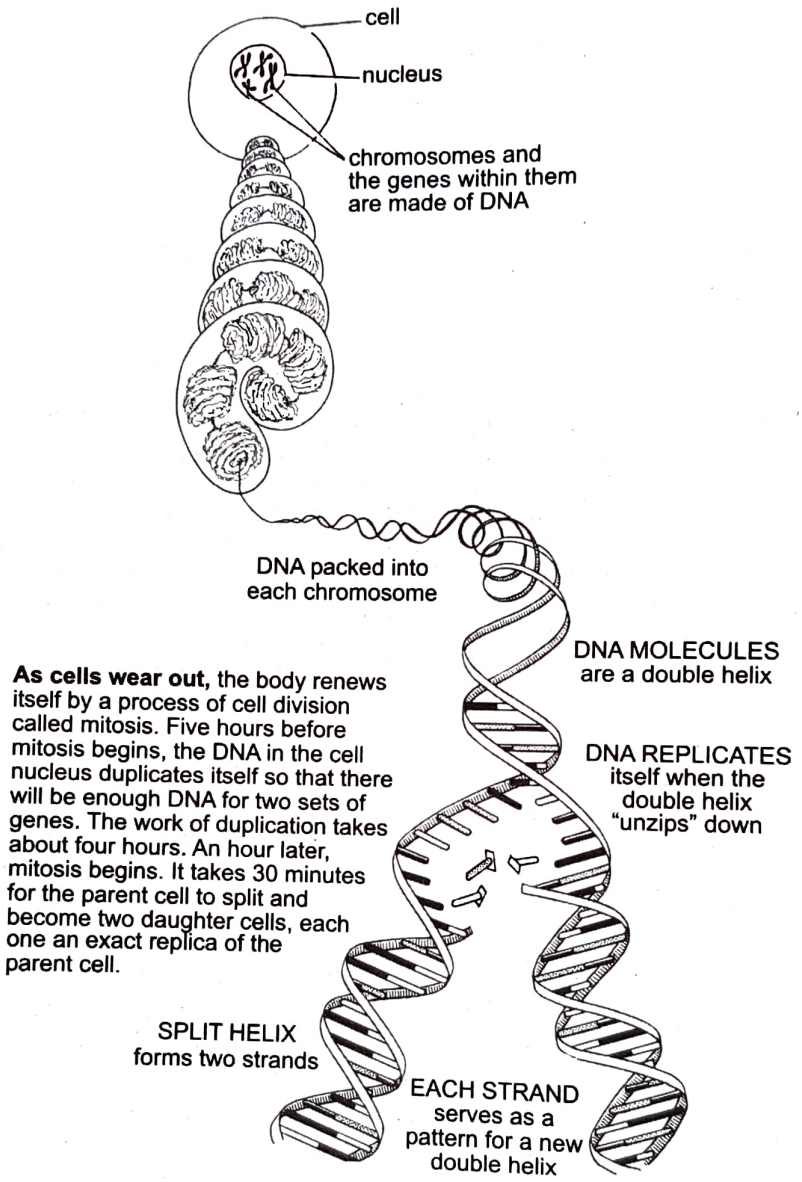


Fig. 1. Genes located on chromosomes constitute total genomic make-up of an organism.

GENES

A gene is a unit of information that directs the activity of the cell or organism during its lifetime. It passes its message along to the progeny when the cell or organism divides or reproduces.

Definition of Gene

In the beginning of the 20th century, Mendel's **factors** (the hereditary units which are transmitted from one generation to other) came to be known as **genes**, a term coined by **W. Johannsen** in 1909. Initially, genes were considered as beads and chromosomes as strings of beads. The gene was regarded as a **unit of function** (cistron), a **unit of mutation** (muton) and a **unit of recombination** (recon). This means that the gene is a hereditary unit, which is independent in controlling a function leading to appearance of a phenotypic trait for which it is meant. It is the smallest

unit that can undergo a mutational change or take part in recombination. This view is based on particulate inheritance, according to which the gene is a locus or a point having no dimensions. Advances in molecular genetics led to fine analysis of the structure and function of the gene and has given a new concept of hereditary unit. With the time, definition of the gene is changed. Some concepts of gene are as follows.

(1) **One gene-one enzyme hypothesis.** The experiments of **George W. Beadle** and **Edward L. Tatum** in the early 1940's on *Neurospora crassa* (the red bread mold) led them to propose one gene-one enzyme hypothesis, which states that one gene controls the production of one enzyme.

(2) **One gene-one polypeptide hypothesis.** Later, it was observed that an enzyme may be composed of more than one polypeptide chain, and a gene can code for only one polypeptide chain. Thus one gene-one polypeptide hypothesis was proposed, that states that one gene controls the production of only one polypeptide chain of an enzyme molecule.

(3) Subsequent studies have shown that besides coding for a polypeptide a gene also codes for the ribosomal and transfer RNAs.

(4) The overlapping genes of certain viruses can code for more than one polypeptide.

(5) Certain genes in some viruses as well as in higher organisms code for a long stretch of a polyprotein.

(6) In eukaryotes, gene expression is not continuous, but it is split into exons and introns and therefore a gene can also contain non-coding sequences.

The above account clearly reveals that though concepts on the structural features of the gene kept on changing with the time, but that **gene is a unit of heredity, is a universal fact.**

Cistron, Recon and Muton

Seymour Benzer (1957) coined the term **cistron, recon** and **muton** as units of **function, recombination** and **mutation** respectively.

1. Cistron. The term **cistron** refers to the continuous segment of DNA which specifies one polypeptide chain. It is the region within which mutants show a **cis-trans position effect**.

2. Muton. A **muton** is the smallest length of DNA capable of giving rise to new form by mutation. It consists of fewer nucleotides than cistron and hence there may be several mutons within a cistron.

3. Recon. A **recon** is the smallest unit that gives rise to new forms by recombination, *i.e.*, it is the shortest distance between two mutons that can allow recombination.

Thus cistron, recon and muton are the three sub-units of the gene in descending order of their size. Sometimes, a recon and a muton may be of the same size.

Properties of Genes (Genetic Material)

The main properties of genes are as follows.

(1) They determine the physical and metabolic characteristics of the cell and are responsible for transmission of characteristics from one cell generation to the other.

(2) They are situated in chromosomes.

(3) There are several genes in each chromosome.

(4) Every gene occupies a fixed position in the chromosome.

(5) They are arranged in a single linear order in a chromosome.

(6) A single gene may occur in several different forms called alleles (multiple allelism). Many genes have only two alleles, each of which controls an alternative expression of the same trait. The two alleles of a gene may frequently be related to each other as dominant and recessive.

(7) A gene may show a sudden change from one form to another. Such a change in form is called mutation and new allele thus formed is known as mutant.

(8) Genes on one chromosome may be transferred to another as a result of crossing over during meiosis or due to translocations.

(9) Duplication of a chromosome during mitosis is preceded by self-duplication of genes of that chromosome.

(10) They express themselves through the production of proteins (enzymes) which control cell metabolism.

Functions of Genes

To qualify to be a gene, it should perform the following three functions.

(1) It should be able to replicate and be inherited by the progeny faithfully.

(2) It should be susceptible to an occasional change by way of mutation and such a change should be stably inherited.

(3) It should be able to carry all the information necessary to programme the functions of a cell.

Nature of Genes

After the establishment of **'gene concept'** biologists concentrated their efforts to find out chemical component of chromosomes which constitutes the actual genetic material. As early as 1869, a Swiss biochemist, **Friedrich Miescher,** from separated nuclei from pus cells isolated a new class of organic compounds which he called **'nuclein'**. It was renamed as **'nucleic acid'** by **Altmann** (1889). By 1920, it became clear that chromosomes are made up of macromolecules of two biological compounds—proteins and deoxyribonucleic acid (DNA). However, there was considerable controversy whether one or both of these constituents are the actual carrier of genetic informations. Proteins being the most complex, their complexity and diversity led many biologists to believe that they are the determinants of diverse characters encountered with in the living world. It was later found that it is not the protein molecule but DNA which is the genetic material. A study of diverse organisms revealed that while nucleic acids are only of two types, there are innumerable kinds of proteins. This created doubt regarding the claim of nucleic acid instead of proteins as the hereditary material. Now, it has been proved beyond doubt that genetic information is stored in nucleic acids of the cell.

Direct evidences for DNA as the genetic material. The most conclusive evidences in support of DNA as the genetic material came from transformation of bacteria and mode of infection of bacteriophages.

Transforming principle (Bacterial transformation experiments). In humans, pneumonia is caused by the bacterium, *Diplococcus pneumoniae,* commonly known as **pneumococcus**. In 1928, **Frederick Griffith**, an English medical bacteriologist, observed two strains of *D. pneumoniae*. In one strain, considerable amount of polysaccharide material is secreted by the cell which forms a large capsule around the cell. The colony produced by these cells has a glistening appearance and is called **smooth (S) type**. In the other strain, no polysaccharide slime layer is secreted by the cell. The colony formed by such cells has an irregular appearance and is said to be **rough (R) type**. The S-strain is **virulent** and can cause pneumonia as the capsule protects the bacteria from mammalian immunologic

defences—phagocytosis by white blood cells. But the non-encapsulated R-strain is **non-virulent** as it can be inactivated by the defense mechanism.

In the course of his work when **Griffith** injected laboratory mice with live R-type pneumococci they suffered no illness. But when the mice were injected with virulent S-type pneumococci, they suffered with pneumonia and died. However, heat killed S-type bacteria did not cause pneumonia.

R-strain → injected into mice → mice live

S-strain → injected into mice → mice die

But when mice were injected with the mixture of living avirulent R-type and heat killed S-type virulent, unexpected symptoms of pneumonia appeared, causing some mice to survive while others died due to pneumonia *(Fig. 2)*. **Griffith** observed that in the blood of dead mice both rough and smooth type of pneumococci occur.

S-strain (heat-killed) [82°C – 90°C] → injected into mice → mice live

S-strain (heat-killed) + R-strain (live) → injected into mice → mice die.

The occurrence of S-type virulent bacteria is possible only by their formation from R-type non-virulent bacteria which pick up the trait of virulent from dead bacteria. The phenomenon is called **Griffith effect** or **transformation**. Griffith proposed that 'transforming principle' is a chemical substance released by heat-killed bacteria. It changed the R-type bacteria to S-type bacteria. It was a permanent genetic change as the new S-type bacteria formed only S-type progeny. He thus concluded that heat-killed smooth type bacteria caused a transformation of the living rough type bacteria. But he could not understand the cause of bacterial transformation.

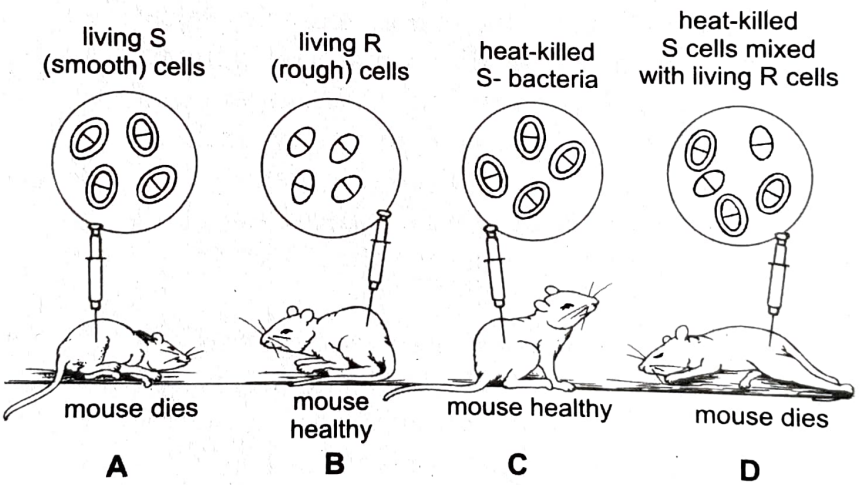


Fig. 2. Griffith's experiment demonstrating transformation in pneumococci.

Transforming substance. Later, **Oswald T. Avery, Colin M. MacLeod** and **Maclyn J. McCarty** in 1944 repeated Griffith's experiments in an *in vitro* system in order to identify the transforming substance responsible for converting non-virulent into virulent type. They found that the DNA isolated from heat-killed S-cells when added to R-cells changed their surface character from rough to smooth and also made them pathogenic *(Fig. 3)*. But when the extract was treated with DNAase (an enzyme which destroys DNA) the transforming ability was lost. Proteases (enzymes which destroy proteins), however, did not affect the transforming ability. These experiments thus suggested that DNA and not proteins is the genetic material. **The phenomenon by which DNA isolated from one type of cell, when introduced into another type, is able to bestow some of the properties of the former to the latter is referred to transformation.**

Conclusion. Griffith's experiment clearly proves that DNA is the store house of genetic information and it acts as genetic material.

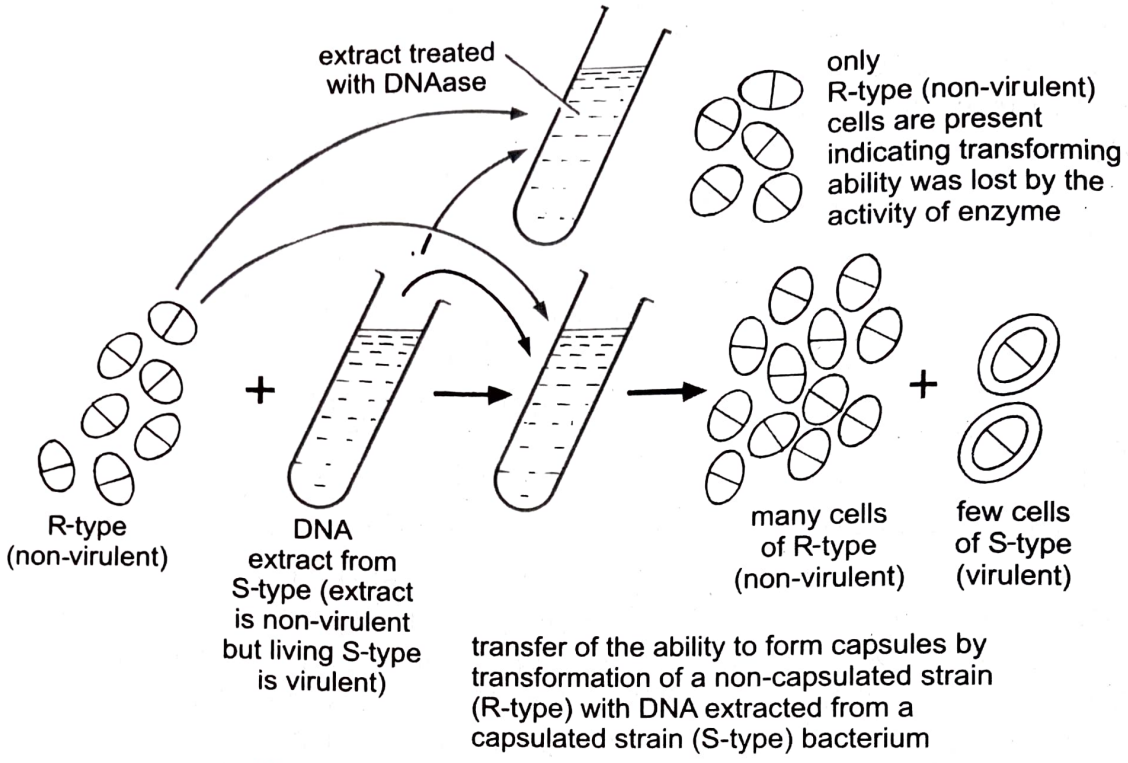


Fig. 3. Transformation experiment of Avery *et al.* (1944).

Evidences from experiments with bacteriophage : Hershey and Chase experiments on T_2 bacteriophage. Many biologists, despite the earlier experiments of **Griffith**, **Avery** and others, still believed that protein, not DNA, is the hereditary material in the cell. As eukaryotic chromosomes consist of roughly equal amounts of protein and DNA, it was said that only protein had sufficient chemical diversity and complexity to encode the information required for genetic material. **In 1952, however, the results of the Hershey-Chase experiment finally provided convincing evidence that DNA is the genetic material.**

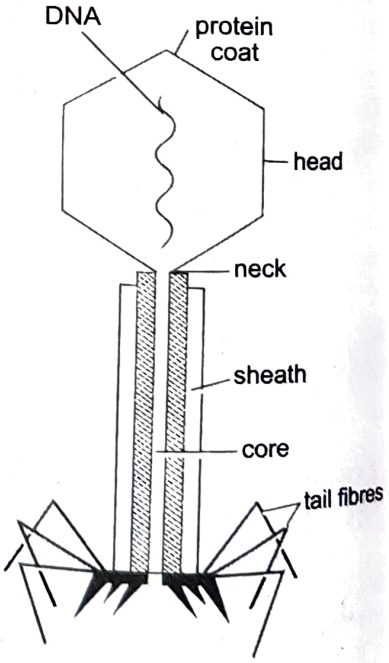


Fig. 4. Structure of T_2 bacteriophage.

Alfred D. Hershey and **Martha Chase** conducted experiments on bacteriophages that attack bacteria. They concentrated on the T_2 bacteriophage that attacks a common bacterium—*Escherichia coli*—in the human digestive tract. This bacteriophage has only two components, protein and DNA. Protein forms the complex external structures (head, sheath, and tail fibres), and a DNA molecule is confined within the head *(Fig. 4)*. The phage attacks *E. coli* by attaching its tail fibres to the bacterial wall. The injecting genetic material that takes over the bacterial metabolic machinery forces it to produce new bacteriophages. At the appropriate time, the bacterium is instructed to produce an enzyme (lysozyme) that ruptures the bacterial cell wall to release hundreds of new bacteriophages.

Hershey and **Chase** realized that this bacteriophage-bacterium system is the ideal material for solving the DNA versus protein controversy. They separately labelled the protein and DNA components of the bacteriophages with specific radioactive tracers and then followed these components through the phages' life cycles.

They developed two strains of viruses, one with labelled proteins and the other with labelled DNA. Almost all proteins contain sulphur, an atom not found in DNA, whereas all DNA molecules contain phosphorus, an atom not found in proteins. Thus, bacteriophages parasitizing bacteria grown in the presence of radioactive sulphur (^{35}S) had labelled proteins, and bacteriophages parasitizing bacteria grown in the presence of radioactive phosphorus (^{32}P) labelled DNA. Such differential labelling thus enables to distinguish between DNA and proteins of the phage.

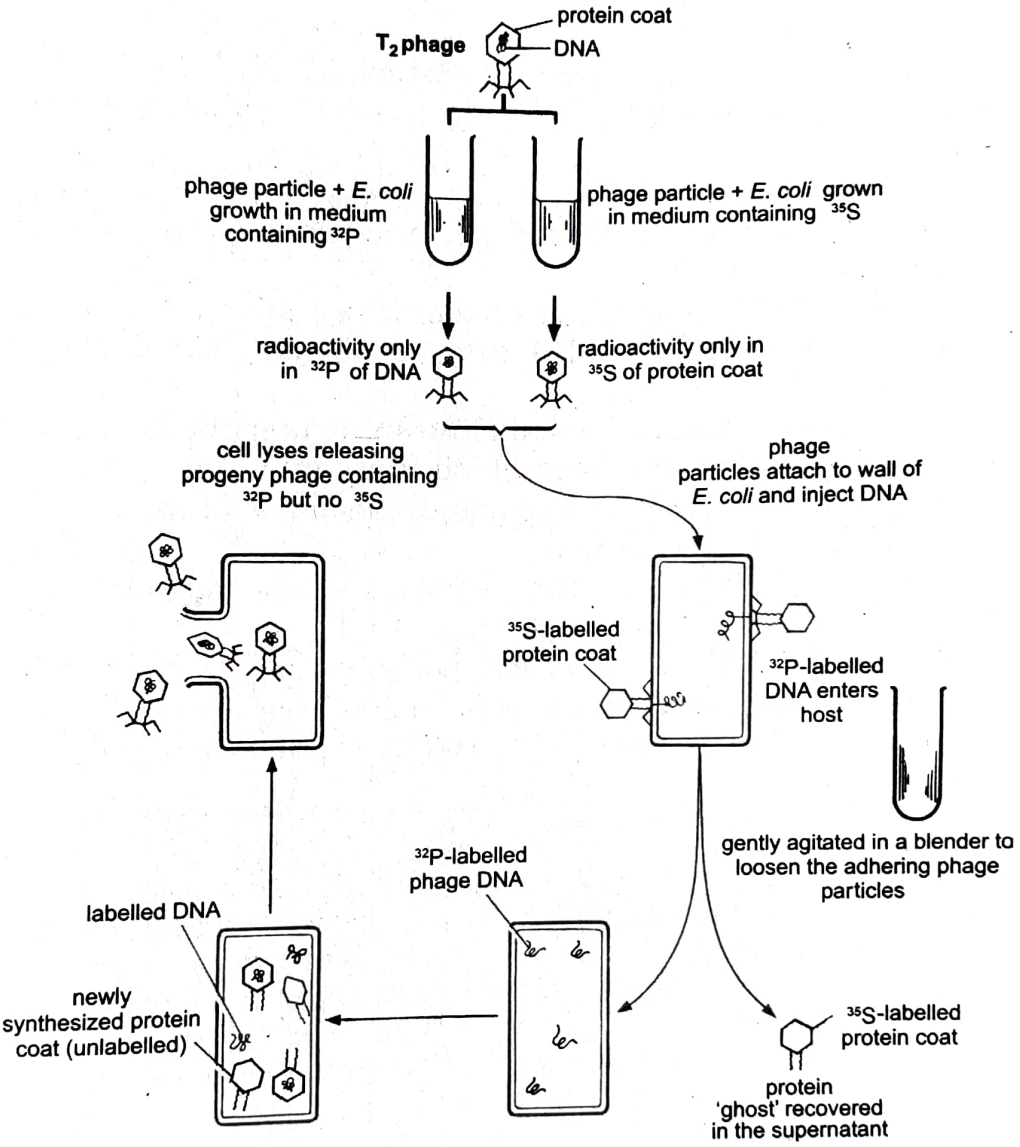


Fig. 5. Hershey and Chase experiment on T_2 bacteriophage.

After developing these two strains, **Hershey** and **Chase** combined each strain with non-radioactive bacteria and allowed bacteriophages to attach and inject their genetic material. Soon after infection (before lysis of bacteria), the bacterial cells were gently agitated in a blender to loosen the adhering phage particles. It was observed that only radioactive ^{32}P was found associated with bacterial cells and ^{35}S was only in the surrounding medium and not in the bacterial cells. When phage progeny was studied for radioactivity, it was found that it carried label only with ^{32}P and not with ^{35}S *(Fig. 5)*. The results clearly indicate that only DNA and not protein coat enters the bacterial cells. It can be thus concluded that it is the viral DNA and not protein that contains information for the production of more viral particles and as such DNA is the genetic material.

Hershey and **Chase** have thus conclusively shown that it is DNA, not protein, that carries the hereditary information.

Properties of genetic material. From the foregoing discussion, it is now clear that it is DNA that acts as genetic material. However, in some viruses like TMV, QB bacteriophage, it is the RNA which acts as the genetic material. A molecule that can act as a genetic material must have the following properties :

(1) It should be able **to replicate**.
(2) It should **chemically and structurally be stable**.
(3) It should be able to express itself in the form of 'Mendelian characters'.
(4) It should be able **to mutate**.

Indirect evidences for DNA as the genetic material. DNA is of universal occurrence in almost all plants and animals. The following are some indirect evidences which suggest that DNA is the genetic material.

(1) It is located on chromosomes.
(2) The quantity of DNA found in a diploid cell is approximately twice of that in a haploid germ cell.
(3) The DNA from cells of widely differing species is less alike in composition than the DNA from closely related species. The DNA from organisms, within the same species has a similar composition.
(4) Among the chemical and physical agents known to alter the chemical structure of DNA without killing the organism, are those which cause mutations.
(5) The wavelengths of U-V light that cause a high incidence of mutation closely correspond to the wavelengths absorbed by DNA.
(6) DNA is stable and its constituent atoms are not exchanged as rapidly as those of other cell molecules.
(7) DNA replicates exactly and exhibits functional specificity.

It can, therefore, be concluded that except in the case of certain viruses (e.g., tobacco mosaic virus, QB bacteriophage), DNA is the substance of which genes of living organisms are composed.

WHICH IS THE GENETIC MATERIAL? DNA *VERSUS* RNA

Hershey-Chase experiment clearly revealed that this is not the protein, but DNA which acts as the genetic material. However, subsequently it is found that in some viruses (e.g., tobacco mosaic virus, QB bacteriophage, etc.), RNA is the genetic material. Later, it is resolved that DNA is the predominant genetic material, whereas RNA performs dynamic functions of messenger. This is ascertained from the differences between chemical structures of DNA and RNA molecules. The genetic material should be stable enough not to change with different stages of life cycle, age or with change in physiology of the organism. Stability as one of the properties of genetic material was very evident in Griffith's 'transforming principle' itself that heat, which killed the bacteria, at least did not destroy the properties of genetic material. In DNA, the two strands being complementary if separated by heating come together, when appropriate conditions are provided. On the other hand, 2′ —OH group present at every nucleotide in RNA is a reactive group and makes RNA labile and easily degradable. Moreover, RNA is reactive due to its catalytic property. Therefore, DNA is less reactive and structurally more stable when compared to RNA. Hence, DNA is a better genetic material.

Furthermore the presence of thymine at the place of uracil also confers additional stability to DNA. Although both DNA and RNA are able to mutate. In fact, RNA being unstable, mutate at a faster rate.

RNA can directly code for the synthesis of proteins, hence can easily express the characters. DNA, however, is dependent on RNA for synthesis of proteins. Thus, the protein synthesising machinery has evolved around RNA. It indicates that both RNA and DNA can function as genetic material, but DNA being more stable is preferred for storage of genetic information. On the other hand, RNA is better for the transmission of genetic information.

Experiments with tobacco mosaic virus (TMV) to demonstrate that RNA is also the genetic material in some cases. H. Fraenkel-Conrat and **B. Singer**, in 1957, first demonstrated that RNA is the genetic material in RNA-containing viruses like TMV *(Fig. 6)*. This virus does not contain any DNA and is composed of RNA (6%) surrounded by a hollow cylinder of protein subunits. **Fraenkel-Conrat** and **Singer** separated RNA from the protein of TMV viruses. Then,

they developed techniques for forming 'reconstituted' viruses containing protein from one mutant strain of TMV and RNA from another, or *vice-versa*. Such hybrid viruses were allowed to infect tobacco leaves, and the progeny were examined. In all cases, the progeny were the parental RNA type and not the parental protein type.

RNA WORLD

A term first used by **Walter Gilbert** in 1986 hypothesizes that RNA was the first genetic material on earth. There is now enough evidence to suggest that essential life processes (such as metabolism, translation, splicing, etc.) evolved around RNA. RNA has the ability to act as both genetic material and as catalyst. There are several biochemical reactions in living systems that are catalysed by RNA (like **ribozymes**). But, RNA being a catalyst was reactive and hence unstable. Hence, it later evolved into a more stable form- the DNA with chemical modifications. Since DNA is a double stranded molecule having complementary strand, it resists changes by evolving a process of repair. These indicate that RNA world may be the original pathway to cells. Recently, **Andrew Z. Fire** and **Craig C. Mellow** (receipient of Nobel Prize in 2006) were of the opinion that RNA is an active ingredient in the chemistry of life.

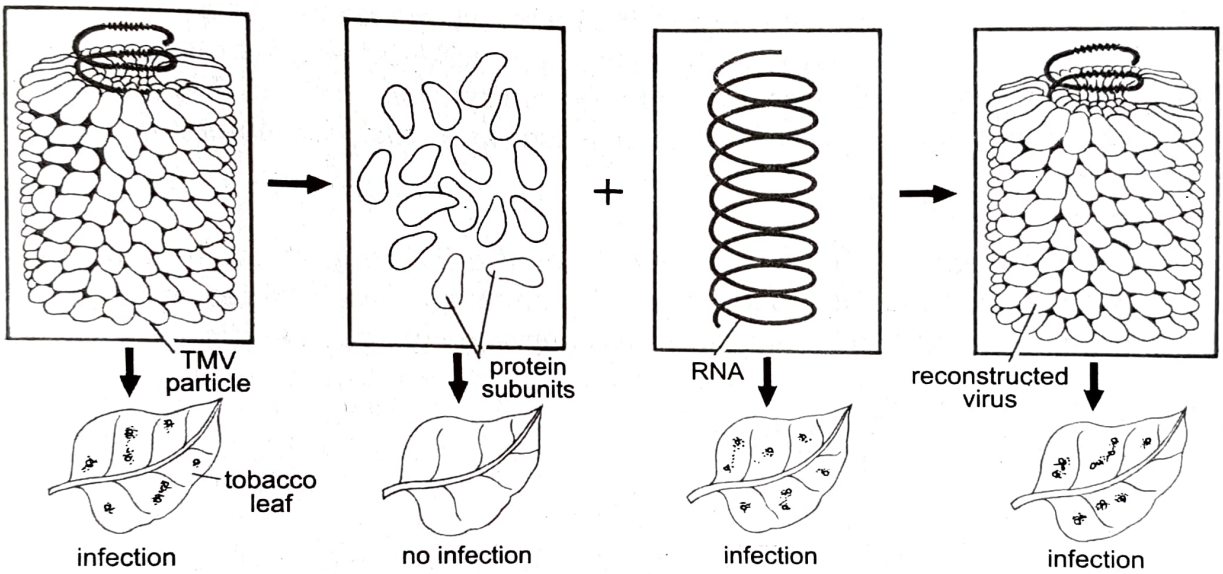


Fig. 6. Experiment of Fraenkel-Conrat on TMV showing that RNA is the genetic material in RNA containing viruses.

NUCLEIC ACIDS

Nucleic acids are the foundation of life as they encode proteins and enzymes which in turn build structural and functional components of the organism. They constitute the genetic material of all living organisms. The information about our bodily features, such as the colour of the skin, hair and eyes, tallness, facial construction, and sex, are in the DNA. DNA analysis is used today to diagnose inherited diseases such as haemophilia, sickle cell anaemia, muscular dystrophy, cystic fibrosis, Alzheimer disease, Huntington's disease, and many others. The knowledge of DNA and the techniques of gene therapy are being used to cure inherited diseases, and to fight against viral and bacterial infections. The DNA fingerprinting, a way of personal identification, which is being used effectively in providing evidences for crimes, settling disputes of parenthood of children, identifying persons in war casualties, etc.

In general, nucleic acids are large polymeric molecules composed of repeating units called nucleotides, which in turn, are composed of three components : a **nitrogenous base** (purine or pyrimidine base), a **pentose sugar**, and **phosphoric acid**. A nitrogenous base is linked to the OH of 1′C pentose sugar through a N-glycosidic linkage to form a **nucleoside** and when a phosphate group is linked to OH of 5′C of a nucleoside through phosphoester linkage, a corresponding **nucleotide** is formed. Two nucleotides are linked through 3′ - 5′ phosphodiester linkage to form

a dinucleotide. More nucleotides are joined in such a manner to form a polynucleotide chain. A polymer thus formed has at one end a free phosphate moiety at 5′ end of sugar, which is referred to as 5′-end of polynucleotide chain. Similarly, at the other end of the polymer the sugar has a free OH of 3′C group which is referred to a 3′-end of the polynucleotide chain. The backbone of a polynucleotide chain is formed due to sugar and phosphates. The nitrogenous bases linked to sugar moiety project from the backbone.

There are two types of nucleic acids, depending on the pentose they contain. Those containing ribose are called **ribonucleic acid** (RNA) and those with deoxyribose are known as **deoxyribonucleic acid** (DNA). DNA is found in chromosomes, in plastids, and in mitochondria. RNA occurs in chromosomes, nucleoli, cytoplasm (as transfer RNA), ribosomes (ribosomal and messenger RNA), plastids and mitochondria. DNA is a blueprint that directs destiny of the cell during its lifetime. It is the most unique molecule in the living world; the only molecule that can replicate itself. It also, in a certain sense, is the material of immortality, since it is the DNA that passes from one generation to the next to maintain genetic continuity between parent and progeny. It is produced mostly in the nucleus but moves out into the cytoplasm. RNA is responsible for transmitting the information from the nucleus to ribosomes where protein synthesis occurs. Besides as a messenger, RNA functions as adapter, structural, and in some cases as a catalytic molecule.

STRUCTURE OF DNA

DNA (deoxyribonucleic acid) is the key molecule of the living system. It is a polynucleotide molecule, consisting of a repeating sequence of monomeric nucleotides arranged in a linear polymeric chain. It is composed of three different chemical substances *(Fig. 7)*.

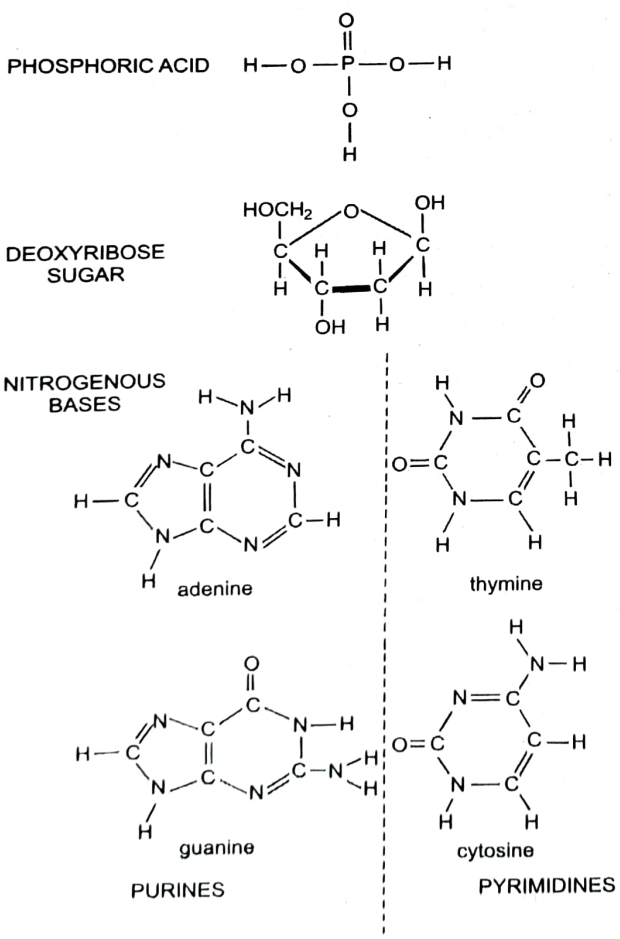


Fig. 7. Building blocks of DNA.

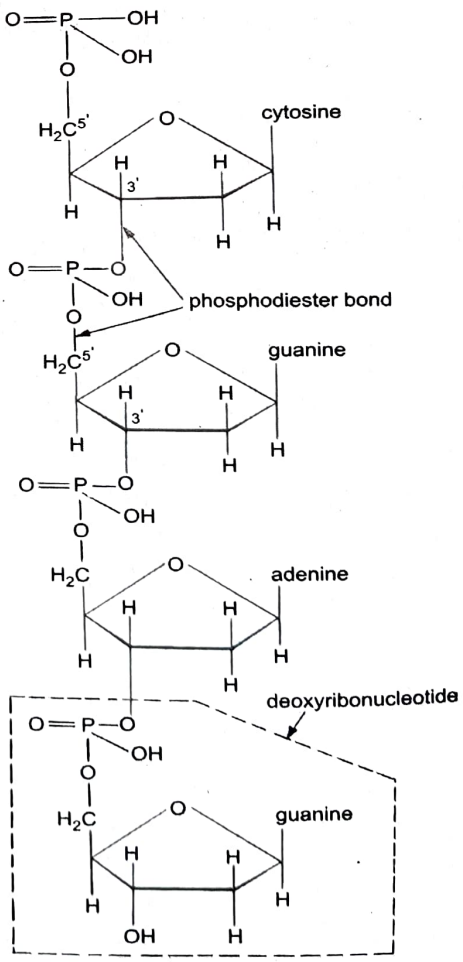


Fig. 8. A polynucleotide of DNA.

(1) A five-carbon (pentose) sugar **deoxyribose**. ($C_5H_{10}O_4$)
(2) A phosphoric acid group. (H_3PO_4)
(3) Usually, four nitrogenous (nitrogen containing) bases—**adenine**, **guanine**, **cytosine** and **thymine**.

Adenine and guanine are **purines**. Purine molecules are double ring structures. These are represented by the letters **A** and **G.** Cytosine and thymine are **pyrimidines**. Pyrimidine molecules consist of a single ring of atoms. These are represented by the letters **C** and **T**.

Table 1. Differences between purines and pyrimidines.

	Purines	Pyrimidines
1.	Adenine and guanine.	Cytosine, thymine, uracil.
2.	9-membered double ringed structures.	6-membered single ringed structures.
3.	Number of carbon atom in the ring is 5.	Number of carbon atom in the ring is 4.
4.	Position of 4 nitrogen atoms are 1, 3, 7 and 9.	Position of 2 nitrogen atoms are 1 and 5.
5.	Large molecular weight.	Relatively low molecular weight.

Chemical analysis of DNA reveals that purine and pyrimidine components occur in equal amounts in a molecule. The total molar amount of adenine in any specimen of DNA is always equal to that of thymine. Thus, the ratio A/T is always one. Similarly, the amount of cytosine is equivalent to that of guanine (Chargaff's rule).

Polynucleotide

A number of nucleotide monomer units may give rise to a polynucleotide chain through the formation of phosphodiester bonds. A phosphodiester bond is formed between any two adjacent nucleotides in which 5′ and 3′ hydroxyls of two adjacent sugars form a double ester with phosphoric acid. At one end of the chain 3′OH group and at the other end 5′OH group is exposed for further elongation of the chain. Thus, each polynucleotide chain has a 3′ and a 5′ end *(Fig. 8)*.

DNA Model as Proposed by Watson and Crick

In 1940, analysis of X-ray diffraction patterns of DNA by **Rosalind Franklin**, **Maurice H.F. Wilkins** and their collaborators was a major step towards the understanding of 3-dimensional structure of DNA. On the basis of X-ray diffraction studies, **M.H.F. Wilkins** (1953) suggested a spiral model of DNA. He noticed that X-ray diffraction patterns of DNAs isolated from varied sources are essentially the same. The important features of DNA structure as revealed by **Franklin** and **Wilkins** are as follows.

(1) It has a helical structure like the bannister of a spiral staircase.
(2) It is long and thin with a uniform diameter of 20 Å.
(3) The distance between successive nucleotides is 3.4 Å.
(4) The helix makes one complete turn every 34 Å long its length.
(5) There are 10 nucleotides per turn of helix.

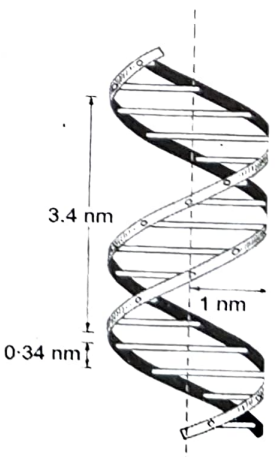


Fig. 9. Features of the DNA molecule as revealed by X-ray diffraction.

With this precise information and application of Chargaff's rule, in 1953, an American geneticist, **James D. Watson** and an English physicist, **Francis H.C. Crick** working together at the Cavendish Laboratory of Cambridge University proposed a model of DNA structure as a **double helix form**. It has proved highly successful both in its ability to account for gene replication and function and in the accuracy of the predictions that can be derived from it. **Watson** and **Crick** realized that if DNA were a single coiled chain of nucleotides positioned 3.4Å apart, with a uniform 20Å diameter, then the molecule would have only half the known density of DNA. Thus, they inferred that in each

DNA molecule there are two long and parallel polynucleotide chains, helically coiled around the same axis. The right-handed helices are held together by their bases which are paired together by covalent hydrogen bonds. Purine of one polynucleotide chain pairs with pyrimidine of the other—adenine with thymine and guanine with cytosine. Both the chains of a DNA molecule are thus complementary to each other. The distance between base pairs is 3.4Å (0.34 nm) and there are ten bases per turn of the double helix. One turn of the helix is as such completed in 34Å. The two helices are coiled in such a way that they can not be separated without unwinding. The backbone of the helix is a chain of sugars and phosphates alternating with each other. The two chains of a helix are of opposite polarity (antiparallel). If one chain runs in 3′ → 5′ direction, the other will run in 5′ → 3′ direction. The width of the helix is 20Å *(Figs. 9, 10)*. For unravelling the structure of DNA, **Watson**, **Crick** and **Wilkins** were awarded Nobel Prize in 1962.

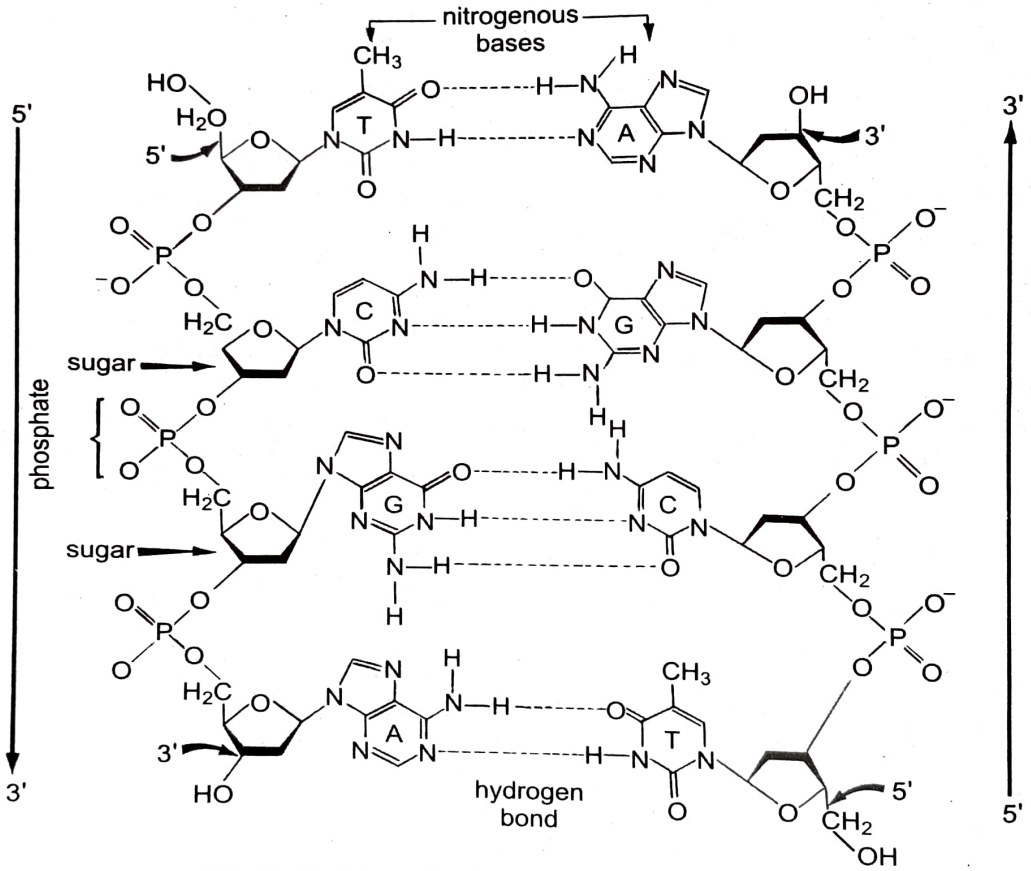


Fig. 10. The Watson-Crick model of the DNA double helix.

The length of DNA is characteristic of an organism. It is usually defined as number of nucleotides (or a pair of nucleotide referred to as base pairs) present in it. For example, $\phi \times 174$ (a bacteriophage) has 5,386 nucleotides, whereas bacteriophage λ (lambda) has 48,502 base pairs (bp), *E. coli* has 4.6×10^6 bp, and of human DNA is 6.6×10^9 bp.

Variants of Double Helix DNA

In nature under physiologic conditions most of the DNA occurs in classic Watson-Crick form which is known as **B-DNA.** It is the most stable structure of DNA molecule. It has 10 base pairs per turn and diameter is 20 nm. It is right handedly coiled. However, X-ray analyses of DNA molecules have revealed that DNA exhibits much more structural diversity and can occur in several variant forms.

1. **A-DNA.** Like B-DNA, A-DNA is a right-handed double helix made of antiparallel strands held together by Watson-Crick base pairing. But the number of base pairs per helical turn is 11, relative to 10.4 base pairs per helical turn found in B-DNA. The helix of A-DNA is wider and shorter than that of B-DNA. In general, in solution, DNA assumes the **B-form** and under conditions of dehydration, the **A-form.** This is because the phosphate groups in the A-DNA bind fewer water molecules than do phosphates in B-DNA.
2. **C-DNA.** C-DNA is also right-handed with 9.33 base pairs per turn of the helix.
3. **D-DNA.** D-DNA is an extremely rare variant with only 8 base pairs per helical turn. Such DNA is devoid of guanine base.
4. **Z-DNA.** Z-DNA was discovered by **Andrew Wang** and **Alexander Rich** in 1979. Unlike other DNAs, Z-DNA is a left-handed double helix with phosphates in the DNA backbone in a *zig-zag* manner. A remarkable characteristic of Z-DNA is that in it the adjacent sugar residues have alternating orientation and it is because of this reason that in Z-DNA, the repeating unit is a **dinucleotide** as against the B-DNA, where the adjacent sugar residues have same orientation so that the repeating unit in B-DNA is a mononucleotide. There are 12 base pairs (six repeating dinucleotide units) per helical turn in Z-DNA. This form occurs mainly in short oligonucleotides that have sequences of alternating pyrimidine and purine bases.

Table 2. Differences in different forms of DNA.

DNA	Name	Handedness of helix	Base pairs per turn	Distance between two base pairs	Stability	Diameter of helix	Length of helix
A	Alternate	Right handed	11	2.5 Å	Unstable	26 Å (widest)	28 Å
B	Balanced	Right handed	10	3.4 Å	Stable and active form	20 Å	34 Å
C	Complementary	Right handed	9.33	3.3 Å	Unstable	19 Å	31 Å
D	Double helix	Right handed	8	3.03 Å	Unstable	19 Å	24 Å
Z	*Zig-zag*	Left handed	12 (6 dimers)	3.8 Å	Unstable	18 Å (thinnest)	46 Å

Table 3. Differences between prokaryotic DNA and eukaryotic DNA.

	Prokaryotic DNA	Eukaryotic DNA
1.	Found in cytoplasm, mitochondria and plastids.	Found in nucleus.
2.	Much less in amount.	Much more in amount.
3.	It is circular.	It is linear.
4.	Naked, without histone proteins.	It is wrapped over histone protein.
5.	Contains only coding regions.	Contains both coding and non-coding regions.
6.	Can code for fewer proteins.	Can code for far more proteins.
7.	G : C contents are more than A : T.	A : T contents are more than G : C.
8.	Repeated sequences absent.	Present.

Single-stranded DNA (ss DNA)

DNA molecules are not always a double-helical structure. Sometimes, it is found as a single-stranded structure. Single-stranded DNA was first observed in $\phi \times 174$ (a virus that infects *E. coli*) by **Robert Sinsheimer** in 1959. ssDNA is somewhat stellate and behaves as a randomly coiled molecule.

STRUCTURE OF RNA

RNA is normally single stranded structure and is composed of a sequence of nucleotides spaced in much the same manner as those of DNA. The nucleotides of RNA consist of bases—adenine (A),

guanine (G), cytosine (C) and uracil (U). Uracil replaces thymine in the RNA molecule. However, just as adenine pairs with thymine in DNA, it pairs with uracil in RNA. The backbone of the nucleic acid is uniformly made up of alternating pentose and phosphate groups. However, in RNA, the pentose sugar is ribose instead of deoxyribose *(Figs. 11, 12)*.

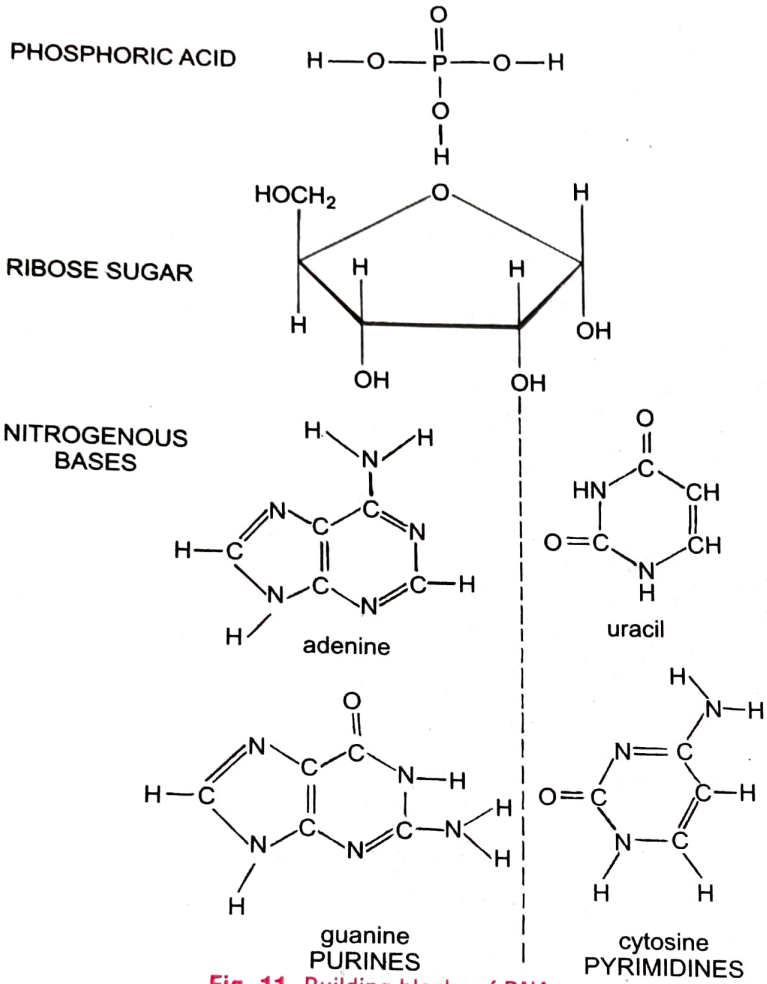


Fig. 11. Building blocks of RNA.

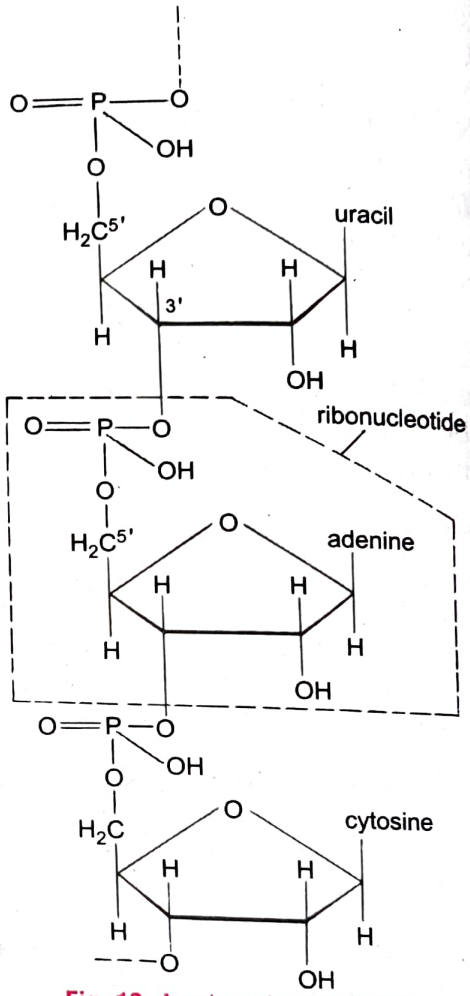


Fig. 12. A polynucleotide of RNA.

Three types of RNA have been identified that vary in size and function. The largest RNA found in ribosomes is known as **ribosomal RNA (rRNA). Messenger RNA** (mRNA) is smaller but is still of considerable size. It is produced in the nucleus and carries the information for the synthesis of proteins. There is a specific mRNA for each protein. **Transfer RNA** (tRNA) is a much smaller molecule and it collects amino acids from the cytoplasm for protein synthesis.

Ribosomal RNA (rRNA). Ribosomal RNA (rRNA) is most abundant of all types of RNAs and makes up about 80% of the total RNA of a cell. It is a single stranded and most stable form of RNA *(Fig. 13)*. It has the highest molecular weight and is sedimented when a cell homogenate containing 10^{-2} M of Mg^{++} is centrifuged at high speed (100,000 gravity for 120 minutes). The unpaired bases in a rRNA molecule binds mRNA and tRNA to ribosomes, possibly by Mg^{++} linkage between phosphate group on the two molecules.

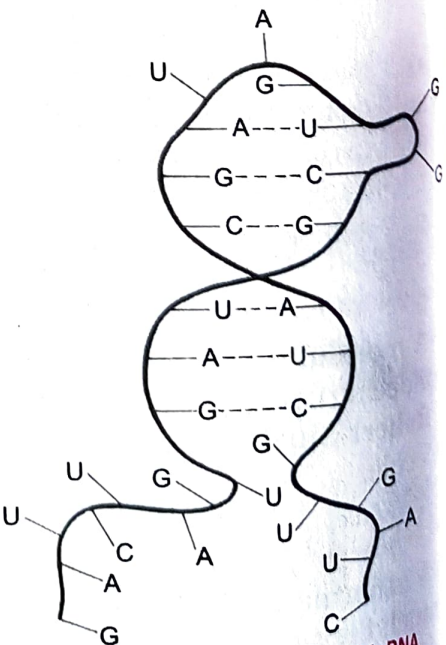


Fig. 13. Molecular structure of rRNA.

It is the structural and functional component of the ribosomes. It is present in the cytoplasm of prokaryotic and eukaryotic cells.

Messenger RNA (mRNA). Template or mRNA is the most heterogeneous in size and stability. It possesses a primary structure similar to a portion of one DNA strand and does not show any secondary or double helical structure at all. It forms a template for protein synthesis. A particular region of the DNA molecule is copied during the synthesis of RNA by replacing thymine with uracil and the sugar is ribose instead of deoxyribose. This copy process involves base pairing properties, *i.e.*, the RNA produced has complementary base to the DNA copied (*i.e.*, U for A, G for C, etc.). The size of mRNA depends on the size of proteins it codes for. In *E. coli*, the average size of mRNA is 900 to 1500 nucleotide units. mRNA carries the coded message from DNA to ribosomes. It directs the amino acid sequence in protein synthesis. It occurs in the nucleolar region as well as in the cytoplasm of prokaryotic and eukaryotic cells. Normally, it carries the codons of single complete protein molecule (**monocistronic mRNA**), but sometimes it carries the codes from several adjacent DNA cistron and becomes much longer in size (**polycistronic mRNA**). In other words, it carries genetic code for proteins. In general, mRNA is short-lived. In bacteria, its half life is of only a few minutes and as such it does not accumulate in the cell.

Table 4. Differences between prokaryotic mRNA and eukaryotic mRNA.

	Characteristics	Prokaryotic mRNA	Eukaryotic mRNA
1.	Nature	Polycistronic (contains information for many proteins)	Monocistronic (contains information only for one protein)
2.	Stability	Less stable	Highly stable
3.	Size	Smaller	Larger
4.	Contains	Only coding regions	Both coding and non-coding regions
5.	Requirement	Post-transcriptional processing not required	Required for functional stability
6.	5 methylated G cap	Absent	Present
7.	Ribosome binding site	Present	Absent
8.	Poly A tail at 3 end	Absent	Present

Transfer RNA (tRNA) : The adapter molecule. The transfer RNA (tRNA) molecules of a cell act as vehicles that pick up amino acids scattered throughout the cytoplasm of prokaryotic and eukaryotic cells and transport them to specific codons of mRNA molecules on ribosomes (i.e., they assist in protein synthesis). It helps to incorporate amino acids into polypeptide chain. Hence, it is also called an **adapter molecule** (a term recognised by **Francis Crick**).

The tRNA molecule consists about 80 nucleotides and has a molecular weight of about 25,000. It is held in a **clover leaf** shape by hydrogen bonds between some of its nitrogenous bases *(Fig. 14)*. In actual structure, the tRNA is a compact molecule which looks like **inverted 'L'**. At one end of the molecule, three unpaired bases form anticodon; they bind with a complementary codon on a mRNA molecule. An anticodon of GAA, for example,

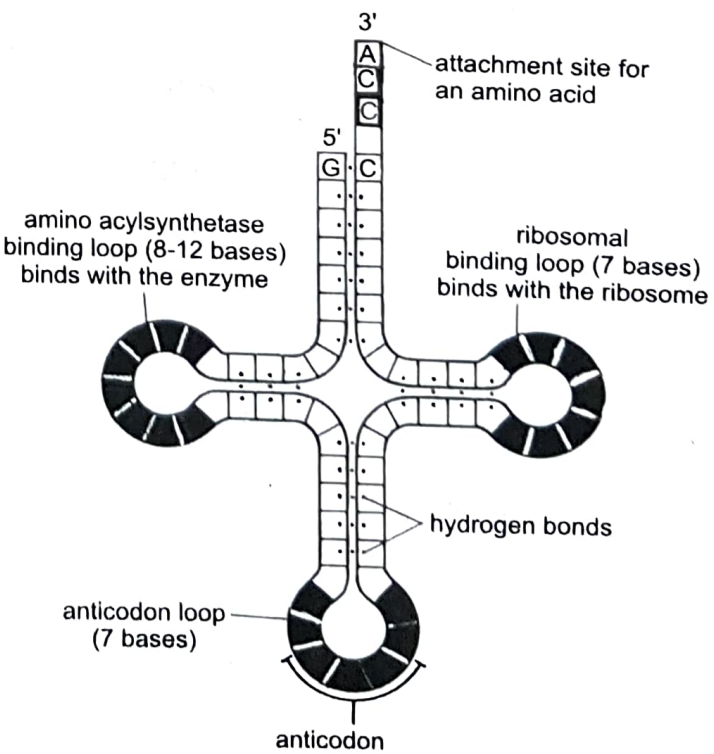


Fig. 14. Clover leaf model of a tRNA molecule to show its basic plan.

forms bonds with a codon of CUU. At the opposite end of the tRNA molecule, its protruding 3' end carries the bases CCA. This is an active site that bonds with the particular kind of amino acid dictated by the anticodon. A tRNA with the anticodon GAA, for example, bonds with and carries only the amino acid leucine. The matching of anticodon with amino acid is done by an enzyme amino acid activating enzyme, or **amino acyl - tRNA synthetase**. There are atleast 20 such enzymes in a cell, one for each kind of amino acid. On one side of a tRNA molecule, there is an active site that binds with the appropriate amino acid activating enzyme, which then facilitates attachment of the appropriate amino acid to the tRNA. The fourth active site is on the side opposite the one that binds with the enzyme. Its function is to recognize a ribosome and by doing so to hold the tRNA to the ribosome in a manner that facilitates codon-anticodon pairing. Each type of tRNA molecule reflects a complementarity in base composition and sequence of its particular gene in the DNA.

These three types of RNA molecules are found in all types of cells. Apart from these RNA there are various other types of RNA molecules found. These are :

- **Small nuclear RNA (sn RNA).** It is a small sized RNA present in the nucleus of eukaryotic cells only. They help in processing of pre-rRNA.
- **Small nucleolar RNA (snoRNA).** It is present in nucleus of eukaryotic cells. They help in processing and assembling rmRNA.
- **Micro RNA (miRNA).** It is present in the cytoplasm of eukaryotic cells. Their function is to inhibit translation of mRNA.
- **Small interfering RNA (si RNA).** Present in the cytoplasm of eukaryotic cells, its function is to trigger degradation of other RNA molecules.

Genomic RNA (Genetic RNA). It is found in riboviruses. It may be single-stranded (e.g. Tobacco mosaic virus) or double-stranded (e.g., Table 5 Retrovirus). It is fragmented in influenza virus. It acts as hereditary material.

Table 5. Differences between DNA and RNA.

	DNA	RNA
1.	Contains deoxyribose sugar.	Contains ribose sugar.
2.	Genetic material.	Not genetic material except in retrovirus.
3.	Double-stranded.	Single-stranded.
4.	Molecular weight very high.	Comparatively low.
5.	Length is very long.	Length is shorter.
6.	It is of only two types-intranuclear and extranuclear.	Many types.
7.	Nitrogeneous base thymine along with adenine, guanine and cytosine.	Thymine is replaced by uracil while other three are similar.
8.	Follows Chargaff rules.	Does not follow.
9.	It replicates to form new DNA molecules.	It cannot normally replicate itself.
10.	DNA transcribes genetic information to RNA.	RNA translates transcribed message for forming polypeptides
11.	Controls metabolism and genetics including variations.	Only controls metabolism under instructions from DNA.
12.	Twisting present.	Usually absent.

Name the following who first:

(a) suggested that an intermediate RNA is required to read the codon on mRNA.

(b) deciphered the genetic code.

(c) used X-rays to cause mutations in *Neurospora*.

(d) suggested inborn errors of metabolism.

(e) proved experimentally that DNA replication is semi-conservative.

(f) proposed double helical structure of DNA.

(g) performed experiments on transformation.

What is genetic material?

Who coined the word gene?

Name the organism in which RNA acts as a genetic material.

Name the scientist who proposed the theory of continuity of germplasm.

Name any two non-sense codons.

Differentiate between codon and anticodon.

What do you understand by central dogma?

If the base sequence of one strand of DNA is CAT, TAG, TAC, GAC, what will be the base sequence of :

(i) Complementary RNA strand

(ii) Complementary RNA strand

Give the schematic diagram to show semi-conservative nature of DNA replication.

What is known by semiconservative nature of DNA?

Distinguish between a leading and lagging strand.

Write the transcribed m-RNA from the DNA strand with the base sequence TAG, TAC, ACT. What is the specific term used for the last codon of the m-RNA in this case?