QP 67019
Answer Key F.Y. B.Sc.
Life Sciences Semester II

Dated: 4.5.2019

Q.1 A. Fill in the blanks **7M**

1. 70S

2. Septate junction

3. Semipermeable

4. Phagocytosis

5. tonofilaments

6. cisternae

7. proteins and peptides

B) Match the columns:
(07)

Column A	**Column B**
a) Photorespiration	i) ATP synthesis
b) mtDNA	ii) Glyoxysomes
c) Phycoerythrin	iii) Amyloplasts
d) Asters	iv) Peoxisomes
e) Starch	v) Rhodoplasts
f) Gluconeogenesis	vi) Petite Yeast
g) Phosphorylation	vii) Centriole

Ans: a)- iv), b)- vi),c)- v), d)- vii), e)- iii), f)- ii), g)- i)

C. Define:

1. Electrophoresis- the movement of charged particles in a fluid or gel under the influence of an electric field.

2. Nitrogenous base- They are particularly important since they make up the building blocks of DNA and RNA: adenine, guanine, cytosine, thymine and uracil.

3. Sphingolipids - any of a class of compounds which are fatty acid derivatives of sphingosine and occur chiefly in the cell membranes of the brain and nervous tissue.

4. Nucleotide- a compound consisting of a nucleoside linked to a phosphate group. Nucleotides form the basic structural unit of nucleic acids such as DNA.

5. Stationary phase in chromatography - Chromatography is used to separate mixtures of substances into their components. ... They all have a **stationary phase** (a solid, or a liquid supported on a solid) and a **mobile phase** (a liquid or a gas). The**mobile phase** flows through the **stationary phase**and carries the components of the mixture with it.

6. RNA - ribonucleic acid, a nucleic acid present in all living cells. Its principal role is to act as a messenger carrying instructions from DNA for controlling the synthesis of proteins, although in some viruses RNA rather than DNA carries the genetic information.

Q.2. A.1. Principle and working of paper chromatography

Paper chromatography is one of the types of chromatography procedures which runs on a piece of specialized paper. It is a planar chromatography system wherein a cellulose filter paper acts as a stationary phase on which the separation of compounds occurs.

Principle of paper chromatography: The principle involved is partition chromatography wherein the substances are distributed or partitioned between liquid phases. One phase is the water, which is held in the pores of the filter paper used; and other is the mobile phase which moves over the paper. The compounds in the mixture get separated due to differences in their affinity towards water (in stationary phase) and mobile phase solvents during the movement of mobile phase under the capillary action of pores in the paper.

The principle can also be adsorption chromatography between solid and liquid phases, wherein the stationary phase is the solid surface of paper and the liquid phase is of mobile phase. But most of the applications of paper chromatography work on the principle of partition chromatography, i.e. partitioned between to liquid phases.

Working:- Chromatography is a method for separating the parts of a mixture of either a gas or liquid solution containing different chemicals. For example, pen inks are often made up of different colours. The different bonding properties of each molecule type is exploited. Chromatography is used in both qualitative and quantitative analysis of both organic and inorganic samples. This technique uses two types of substances:

1. Mobile phase: a gas or liquid that transports the solution being tested through the other substance (water, rubbing alcohol are examples).
2. Stationary phase: the liquid or solid through which the tested substance is carried (coffee filter paper, paper towel are examples).

It is necessary for the different chemicals in the solution fo have different properties such as molecule size or a different ability to dissolve in a solvent. The stationary phase will absorb or slow down different components of the tested solution to different degrees creating layers as the components of the solution are separated. Chromatography was invented by the Russian botanist, Mikhail Tsvet. Chemists use this process to identify unknown substances by separating them into the different molecules that make them up.

Uses and Applications of Paper Chromatography

Paper chromatography is specially used for the separation of a mixture having polar and non-polar compounds.

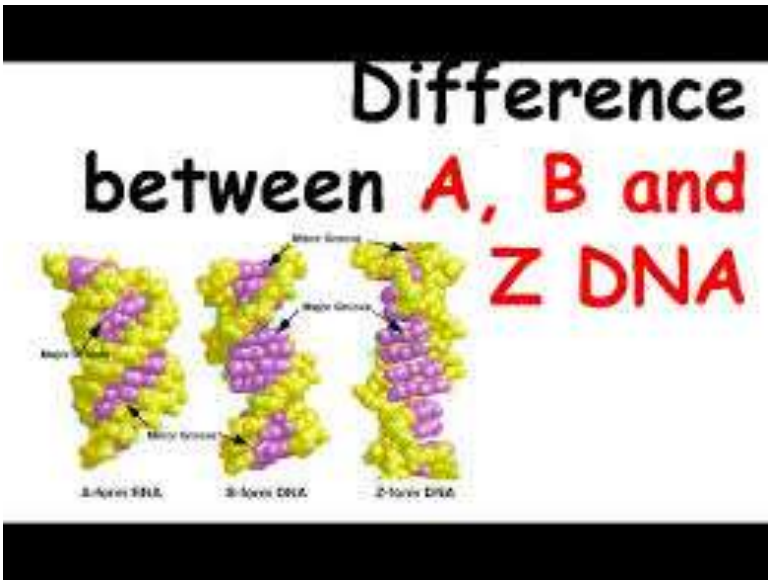
For separation of amino acids.

It is used to determine organic compounds, biochemicals in urine, etc.

In the pharma sector it is used for the determination of hormones, drugs, etc.

Sometimes it is used for evaluation of inorganic compounds like salts and complexes

Q.2. A. 2. Structure B-DNA. Difference with A and Z forms of DNA



DNA in the cell functions as a double-stranded helix of B-form DNA, the structure of which was first determined by Watson and Crick. Here the two strands are differentially colored to illustrate the right handed B-form helix clearly.

The polar sugar-phosphate backbones of each strand form the helical scaffold, with the nitrogenous bases in the interior of the molecule, their planes nearly perpendicular to the helical axis. Each base forms hydrogen bonds (indicated by dashed lines) with a base from the opposite strand.

The polarity of the backbones is antiparallel, with one strand running *3′* ---> *5′* and the other*5′* --->*3′*. This can readily be seen by observing the reversed orientations of the ribose sugars on opposite strands.

The diameter of the B-DNA is ~20 Angstroms, and the distance between base pairs is ~3.4 Angstroms.

The base pairing of opposite strands is stereochemically selective, Adenine always pairing with Thymine, and Guanine with Cytosine. Two and three hydrogen bonds are formed in A-T and G-C base pairs, respectively.

A-T and G-C base pairing results in strand complementarity, with one strand of the double helix forming a sequence of bases complementary in hydrogen bonding to that of the other strand. As noted by Watson and Crick in their 1953 paper describing the first model of the DNA double helix, base complementarity provides a means by which the genetic material can replicate with fidelity. Each strand of a parent double helix serves as a template upon which

complementary strands are built, producing two daughter molecules identical to the parent, with one strand of each daughter helix conserved from the parent molecule.

This semi-conservative mode of replication is here illustrated schematically for 1 strand of DNA.

The attachment of bases to the backbone sugars through glycosidic bonds is asymmetrical. This results in the formation of two different grooves on opposite sides of the base pairs, the major and minor grooves. Although the grooves are of similar depth in B-DNA, the major groove is considerably wider than the minor groove.

The edges of the base pairs present a more complex sterochemical environment in the major groove than in the minor groove. As can be observed in the T-A base pair shown, the major groove edge contains a methyl group (CH3), a hydrogen bond acceptor (HA), a hydrogen bond donor (HD), and a second hydrogen bond acceptor (HA) - for simplicity, hydrogens are not shown. The minor groove edge, in contrast, presents only a HA, a hydrogen (H), and a second HA. Whereas a switch to an A-T base pair would present a reversed edge chemistry in the major groove (HA-HD-HA-CH3), the edge in the minor groove would remain the same as in a T-A pair (HA-H-HA). Similar considerations apply to C-G and G-C base pairs. Since many proteins that bind DNA recognize specific sequences of bases, it is not surprising that most bind to the floor of the major groove, as this provides more chemical information for recognition than the minor groove.

The distance between the sugar-phosphate backbones is greater in the major groove than in the minor groove.

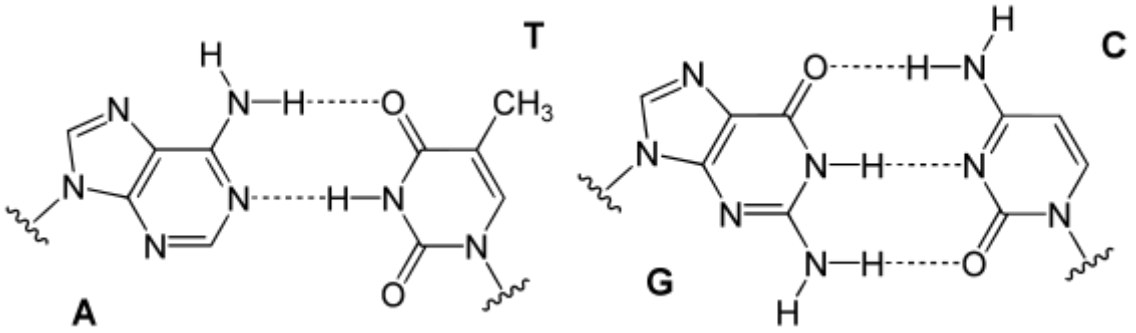
The major and minor grooves lie 180^{o}opposite each other in the double helix, spiraling along the axis of the molecule.

Three major forms of DNA are double stranded and connected by interactions between complementary base pairs. These are terms A-form, B-form,and Z-form DNA.

B-form DNA

The information from the base composition of DNA, the knowledge of dinucleotide structure, and the insight that the X-ray crystallography suggested a helical periodicity were combined by Watson and Crick in 1953 in their proposed model for a double helical structure for DNA. They proposed two strands of DNA -- each in a right-hand helix -- wound around the same axis. The two strands are held together by H-bonding between the bases (in anti conformation) as shown in Figure 2.5.12.5.1.

Major groove Major groove



Minor groove Minor groove

Figure 2.5.12.5.1***:*** *(left) An A:T base pair and (right) a G:C base pair*

Bases fit in the double helical model if pyrimidine on one strand is always paired with purine on the other. From **Chargaff's rules**, the two strands will pair A with T and G with C. This pairs a keto base with an amino base, a purine with a pyrimidine. Two H-bonds can form between A and T, and three can form between G and C. This third H-bond in the G:C base pair is between the additional exocyclic amino group on G and the C2 keto group on C. The pyrimidine C2 keto group is not involved in hydrogen bonding in the A:T base pair.

These are the complementary base pairs. The base-pairing scheme immediately suggests a way to replicate and copy the the genetic information.

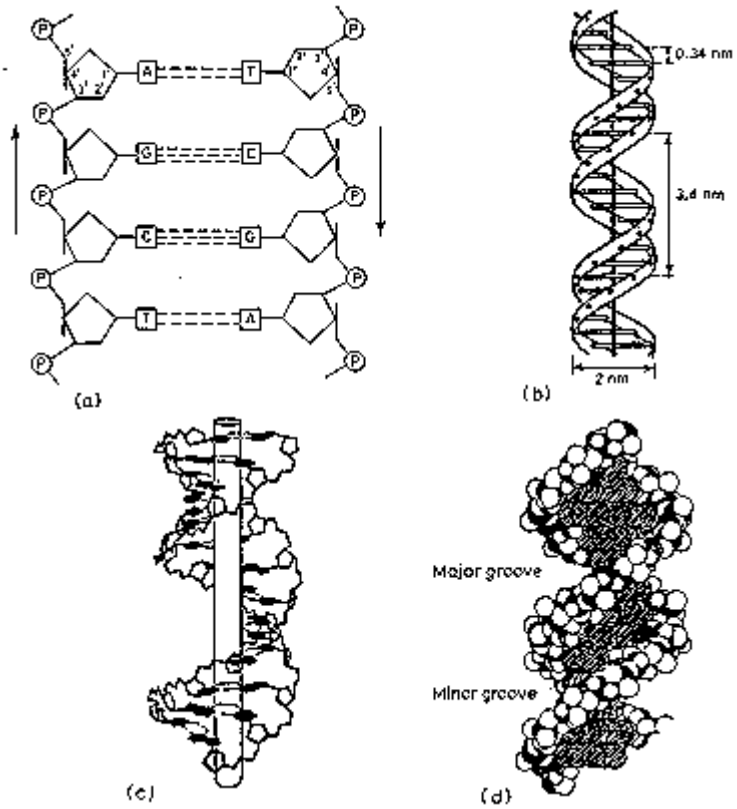
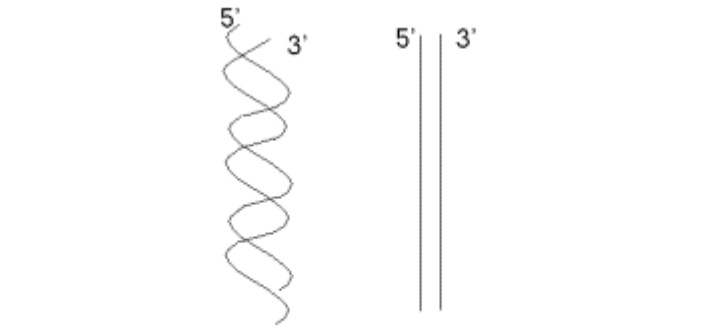


Figure 2.5.22.5.2***:*** *Antiparallel (a), plectonemically coiled (b, c, d) DNA strands. The arrows in a are pointed 3' to 5', but they illustrate the antiparallel nature of the duplex. The two strands of the duplex are antiparallel and plectonemically coiled. The nucleotides arrayed in a 5' to 3' orientation on one strand align with complementary nucleotides in the the 3' to 5' orientation of the opposite strand.*

The two strands are not in a simple side-by-side arrangement, which would be called a *paranemic joint* (Figure 2.5.32.5.3). (This will be encountered during recombination in Chapter 8.) Rather the two strands are coiled around the same helical axis and are intertwined with themselves (which is referred to as a **plectonemic coil**). One consequence of this intertwining is that the two strands cannot be separated without the DNA rotating, one turn of the DNA for every "untwisting" of the two strands.



In a plectonemic coil, the two strands wrap around each other.
In a paranemic joint, the two strands align side-by-side.

Figure 2.5.32.5.3***:*** *Duplex DNA has the two strands wrapped around each other in a plectonemic coil (left), not a paranemic duplex (right).*

Dimensions of B-form (the most common) of DNA

- 0.34 nm between bp, 3.4 nm per turn, about 10 bp per turn
- 1.9 nm (about 2.0 nm or 20 Angstroms) in diameter

Major and minor groove

The major groove is wider than the minor groove in DNA (Figure 2.5.2d2.5.2d), and many sequence specific proteins interact in the major groove. The N7 and C6 groups of purines and the C4 and C5 groups of pyrimidines face into the major groove, thus they can make specific contacts with amino acids in DNA-binding proteins. Thus specific amino acids serve as H-bond donors and acceptors to form H-bonds with specific nucleotides in the DNA. H-bond donors and acceptors are also in the minor groove, and indeed some proteins bind specifically in the minor groove. Base pairs stack, with some rotation between them.

A-form nucleic acids and Z-DNA

Three different forms of duplex nucleic acid have been described. The most common form, present in most DNA at neutral pH and physiological salt concentrations, is B-form. That is the classic, right-handed double helical structure we have been discussing. A thicker right-handed duplex with a shorter distance between the base pairs has been described for RNA-DNA duplexes and RNA-RNA duplexes. This is called A-form nucleic acid.

A third form of duplex DNA has a strikingly different, left-handed helical structure. This Z DNA is formed by stretches of alternating purines and pyrimidines, e.g. GCGCGC, especially in negatively supercoiled DNA. A small amount of the DNA in a cell exists in the Z form. It has been tantalizing to propose that this different structure is involved in some way in regulation of some cellular function, such as transcription or regulation, but conclusive evidence for or against this proposal is not available yet.

Differences between A-form and B-form nucleic acid

The major difference between A-form and B-form nucleic acid is in the conformation of the deoxyribose sugar ring. It is in the C2' endoconformation for B-form, whereas it is in the C3' endoconformation in A-form. As shown in Figure 2.5.42.5.4, if you consider the plane defined by the C4'-O-C1' atoms of the deoxyribose, in the C2' endoconformation, the C2' atom is above the plane, whereas the C3' atom is above the plane in the C3' endoconformation. The latter conformation brings the 5' and 3' hydroxyls (both esterified to the phosphates linking to the next nucleotides) closer together than is seen in the C2' endoconfromation (Figure 2.16). Thus the distance between adjacent nucleotides is reduced by about 1 Angstrom in A-form relative to B-form nucleic acid (Figure 2.5.42.5.4).

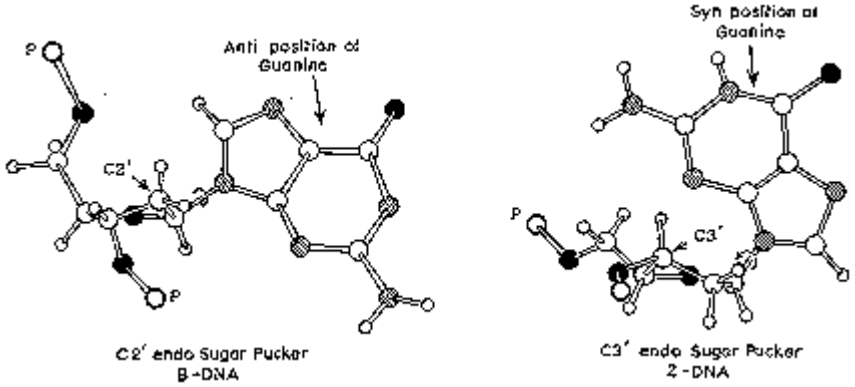


Figure 2.5.42.5.4***:*** *Syn and anti conformations of the base relative to the sugar in nucleotides.*

A second major difference between A-form and B-form nucleic acid is the placement of base-pairs within the duplex. In B-form, the base-pairs are almost centered over the helical axis (Figure 2.5.42.5.4), but in A-form, they are displaced away from the central axis and closer to the major groove. The result is a ribbon-like helix with a more open cylindrical core in A-form.

Z-form DNA

Z-DNA is a radically different duplex structure, with the two strands coiling in left-handed helices and a pronounced zig-zag (hence the name) pattern in the phosphodiester backbone. As previously mentioned, Z-DNA can form when the DNA is in an alternating purine-pyrimidine sequence such as GCGCGC, and indeed the G and C nucleotides are in different conformations, leading to the zig-zag pattern. The big difference is at the G nucleotide. It has the sugar in the C3' endoconformation (like A-form nucleic acid, and in contrast to B-form DNA) and the guanine base is in the synconformation. This places the guanine back over the sugar ring, in contrast to the usual anticonformation seen in A- and B-form nucleic acid. Note that having the base in the anticonformation places it in the position where it can readily form H-bonds with the complementary base on the opposite strand. The duplex in Z-DNA has to accomodate the distortion of this G nucleotide in the synconformation. The cytosine in the adjacent nucleotide of Z-DNA is in the "normal" C2' endo, anticonformation.

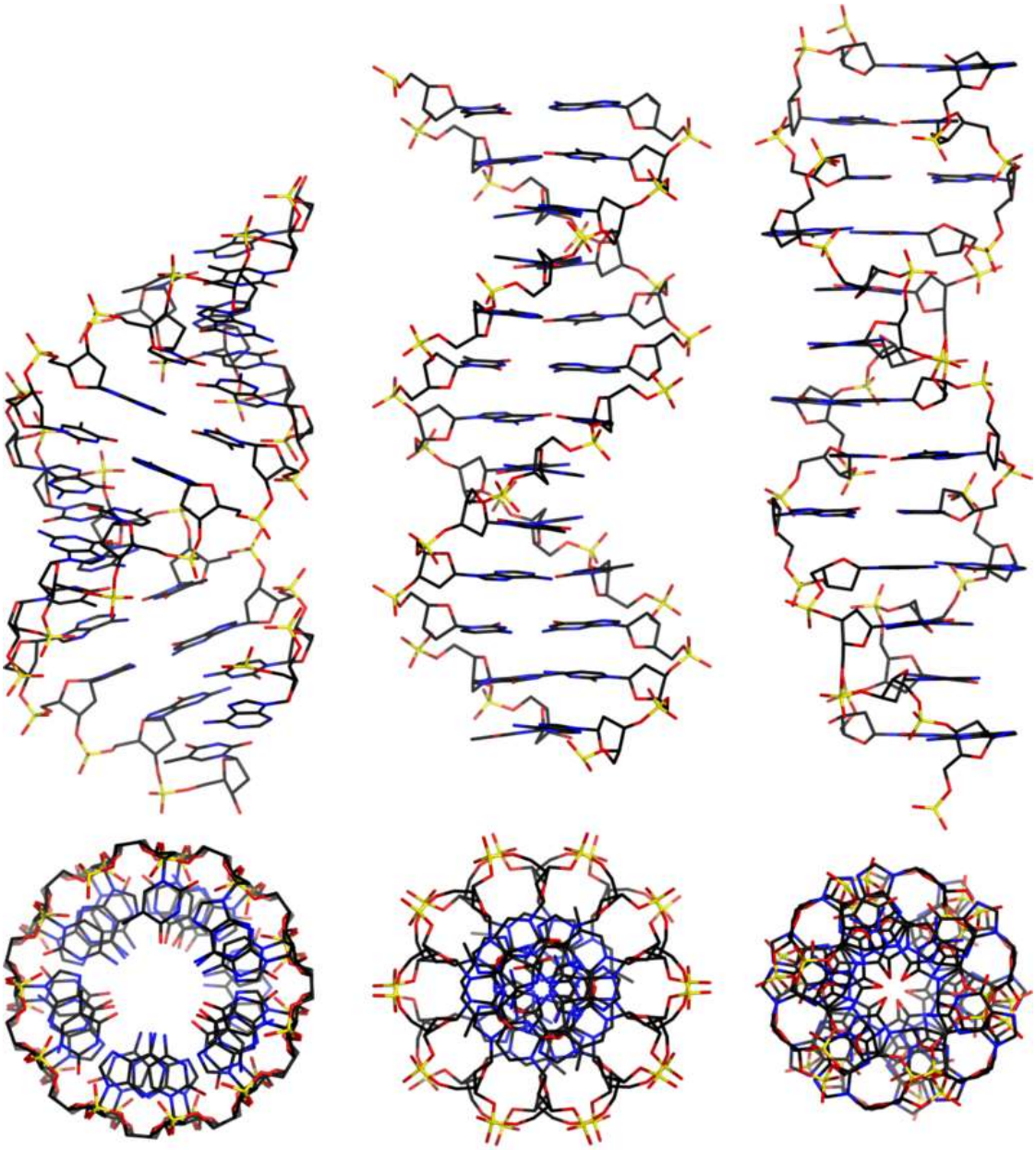


Figure 2.5.52.5.5***:*** *B-form (left), A-form (middle) and Z-DNA (right). Image used with permission (CC BY-SA 4.0; Mauroesguerroto)*

Even classic B-DNA is not completely uniform in its structure. X-ray diffraction analysis of crystals of duplex oligonucleotides shows that a given sequence will adopt a distinctive structure. These variations in B-DNA may differ in the propeller twist (between bases within a pair) to optimize base stacking, or in the 3 ways that 2 successive base pairs can move relative to each other: twist, roll, or slide.

Table 2.5.12.5.1 *Comparisons of B-form, A-form and Z-DNA*

	B-Form	A-Form
helix sense	Right Handed	Right Handed
base pairs per turn	10	11
vertical rise per bp	3.4 Å	2.56 Å
rotation per bp	+36°	+33°
helical diameter	19 Å	19 Å

Q. 2. B.1. Fractionation by ammonium sulphate

Ammonium sulphate fractionation

Proteins are soluble in aqueous media because they have hydrophilic amino acid side-chains facing outwards that can interact with water. These are provided by the basic amino acids (arginine, histidine, arginine and lysine), the acidic amino acids (aspartate and glutamate) and the neutral hydrophilic amino acids (asparagine, glutamine, serine, threonine, tyrosine and cysteine).

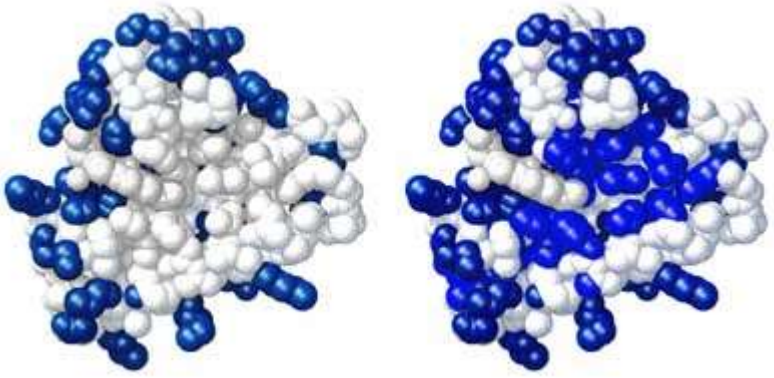
Any compound that interferes with these interactions between amino acid side-chains and water, by reducing the available water, will reduce the solubility of the protein. As interactions with water become less marked, so protein-protein interactions become more important, and the protein will aggregate and come out of solution. Provided that the temperature is maintained low enough (around 4C), the protein is not irreversibly denatured, but the precipitate can be redissolved in buffer.

A number of different methods can be used to reduce hydrophilic interactions and precipitate out proteins reasonably selectively, including:

- Salting out with ammonium sulphate

- Selective precipitation with an organic solvent that both reduces available water and also decreases the dielectric constant of the solution. Ethanol fractionation is commonly used to separate protein fractions from blood plasma. Historically, acetone precipitation was an important way of obtaining a dry powder that could be stored for some time without losing activity;
- Selective precipitation using non-ionic polymers such as polyethylene glycol, which both reduce the available water and may also interact directly with some of the proteins in the mixture, in much the same way as do the polymers used in gel filtration.

Ammonium sulphate is highly hydrated, and a concentrated ammonium sulphate solution reduces the available water very considerably.



The diagram on the right shows two proteins, with their hydrophilic regions coloured blue.The protein on the left has relatively few hydrophilic regions, and hence will aggregate and precipitate at a relatively low concentration of ammonium sulphate - perhaps around 20 - 30% saturation. By contrast, the protein on the right has considerably more hydrophilic regions, and hence will remain in solution until the concentration of ammonium sulphate is considerably higher - perhaps around 50 - 60% saturation.

This means that it is possible to separate proteins from a mixture on the basis of their relative hydrophilicity by gradually increasing the concentration of ammonium sulphate.

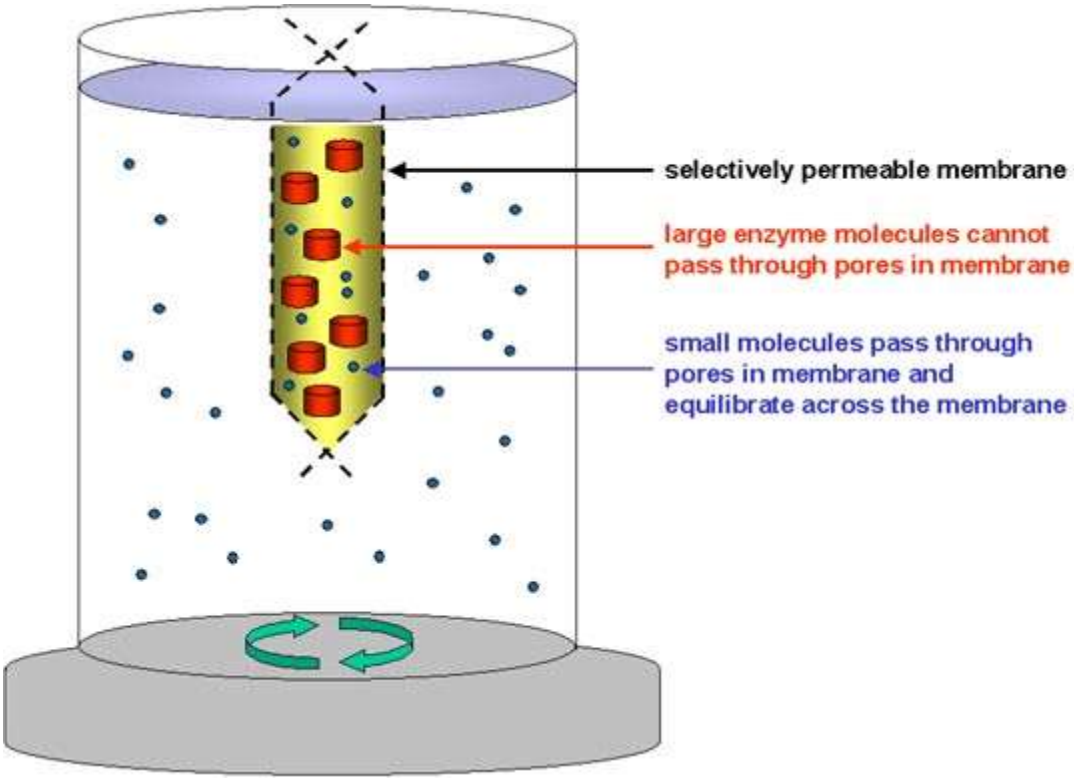
At each stage you calculate the volume of saturated ammonium sulphate solution that will be required to achieve a given percentage saturation of your enzyme preparation, which is typically a crude tissue homogenate or perhaps a high-speed supernatant of a tissue homogenate. The ice-cold saturated solution of ammonium sulphate is added slowly to the protein solution, in an ice bath, and stirred continually. When the required amount has been added, the solution is centrifuged, and the precipitate collected, and redissolved in buffer. A higher degree of saturation with ammonium sulphate is then achieved by adding more saturated ammonium sulphate solution in the same way.

Initially you would probably use a number of wide ranges of ammonium sulphate saturation, say 0 - 50% and see whether or not your enzyme is precipitated. If it is, then you can refine the range until you achieve maximum recovery of the enzyme and maximum removal of interfering proteins.

Removing the ammonium sulphate by dialysis

Having precipitated a protein fraction that contains most of your enzyme, and redissolved it in buffer, it is necessary to remove the ammonium sulphate before you can proceed to subsequent steps in the purification process. The simplest way to achieve this is to dialyse the

solution.

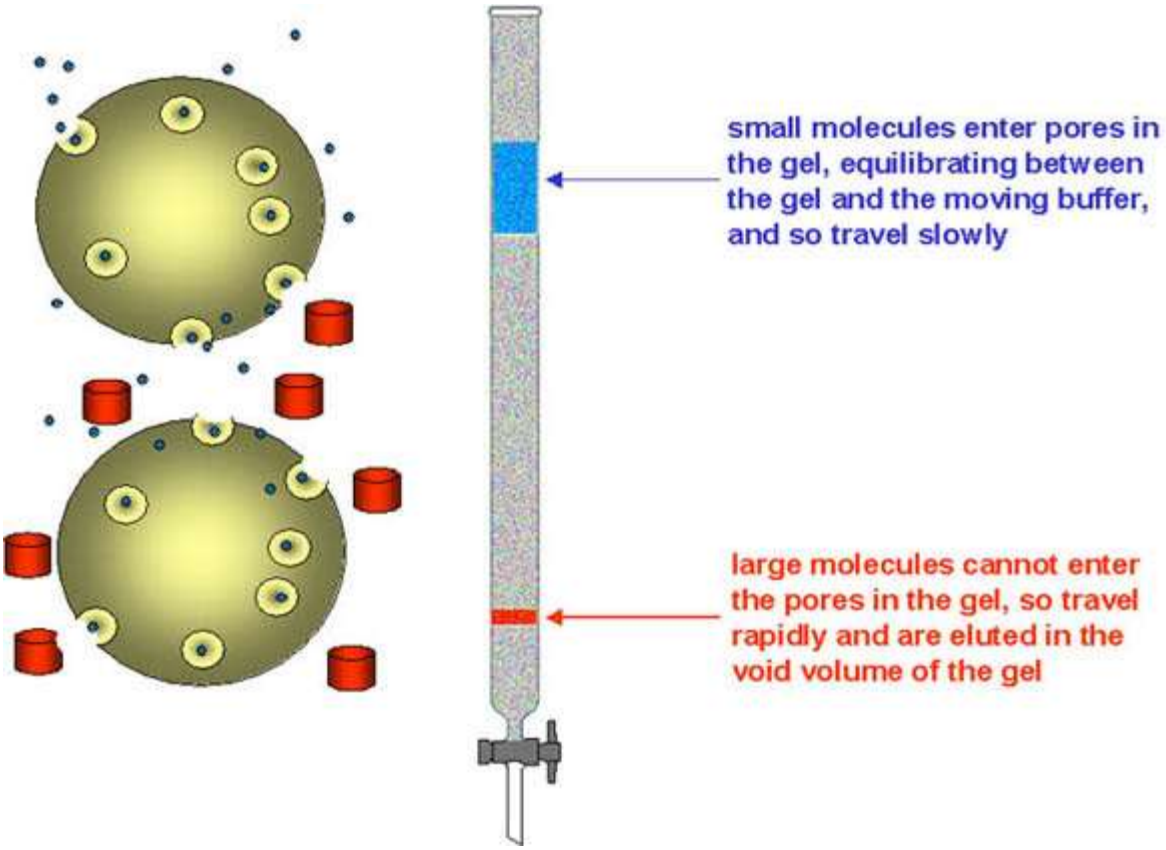


As shown in the diagram, the enzyme solution is placed in a bag of selectively permeable membrane (e.g. cellophane), immersed in a large volume of buffer that is stirred and maintained at about 4C.The membrane has pores that will permit small molecules such as ammonium and sulphate ions to cross, and hence equilibrate in the larger volume of buffer outside, while not permitting large protein molecules to cross. If the buffer is changed several times, allowing several hours each time for the ammonium sulphate to equilibrate, more or less all of it will be removed from the protein solution.

Dialysis will increase the volume of the enzyme solution, because of the initial osmotic effect of the ammonium sulphate; (this is why it is important to leave an air gap at the top of the membrane tube, to prevent it bursting).

Removing the ammonium sulphate by gel filtration

An alternative way of removing the ammonium sulphate is by gel filtration, using e.g. Sephadex G25, which has small pores that will retard small molecules such as ammonium and sulphate ions, but will exclude large protein molecules, so that they are eluted in the void volume of the column. This means that the early eluate will contain the proteins, more or less free from ammonium sulphate.



2. Diagram of double helical structure of DNA

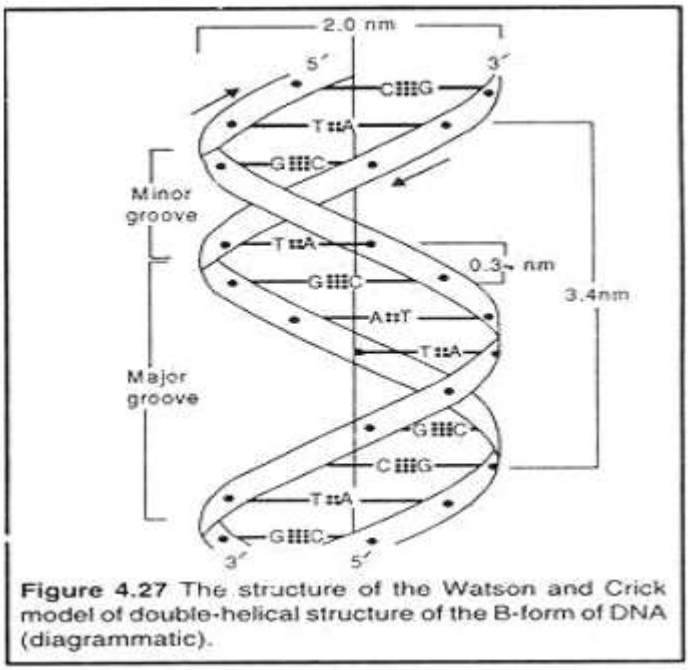


Figure 4.27 The structure of the Watson and Crick model of double-helical structure of the B-form of DNA (diagrammatic).

3. Differentiate between DNA and RNA

Table 5.1. Difference between RNA and DNA

RNA	DNA
RNA is single stranded except in some viruses	DNA is double stranded except in few viruses
RNA have ribose sugar	DNA have deoxyribose sugar
Bases present are adenine, guanine, cytosine and uracil.	Bases present are adenine, guanine, cytosine and thymine.
Adenine pairs with uracil	Adenine pairs with thymine
Purine is not equal to pyrimidine	Purine is equal to pyrimidine (Chargaff's rule)
Regions having complementary nucleotides, pairs, and form hair pin loop like structure and helical.	Complementary nucleotides are present throughout the length of the DNA.
RNA is genetic material in some viruses.	DNA is the genetic material in all living organisms.
Length of RNA is short consisting of only few thousands nucleotides.	Length of DNA is quite large consisting of millions of nucleotides.
Three types of RNA are present in an organism: mRNA, rRNA, tRNA.	DNA occurs only in one form in an organism.
mRNA occurs in nucleolus, rRNA and tRNA occur in cytoplasm.	DNA occurs in nucleus, nucleolus, and extrachromosomal DNA in mitochondria and chloroplast.

4. Biological functions of lipids

Lipids can serve a diverse range of functions within a cell, including:

- **Storage of energy** for long-term use (e.g. triglycerides)
- **Hormonal roles** (e.g. steroids such as oestrogen and testosterone)
- **Insulation** – both thermal (triglycerides) and electrical (sphingolipids)
- **Protection** of internal organs (e.g. triglycerides and waxes)
- **Structural components** of cells (e.g. phospholipids and cholesterol)

Q.3.A.Describe any 1 **10M**

1. Polymorphism of lysosomes—

Describe formation of primary lysosomes, heterophagosomes, Autophagosomes and residual bodies

Functions of lysosomes—

Digestion of large extracellular particles

Digestion of intracellular substances

Autolysis

Extracellular digestion

2. Structure and biochemical composition of 70S and 80S ribosomes

Formation of 70S ribosome from 50S and 30S two subunits

Formation of 80S ribosome from 60S and 40S two subunits

Large subunit is dome shaped and smaller subunit forms cap like structure.

The binding of two subunits is dependent on Mg ions concentrations

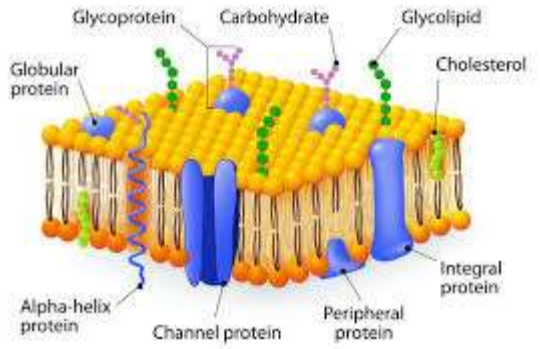
Composition 70S small subunit made of 16rRNA plus 21 proteins

And large subunit made of 23S, 5SrRNAs and 34 proteins

80S small subunit is made of 18SrRNA and 30 proteins and larger subunit has 28S, 5.8S and 5S rRNAs and 40 proteins

Q.3. B Explain any 2 **10M**

1. Fluid mosaic model



2. Golgi apparatus functions in Plants—

Secretion of materials of primary and secondary cell walls, formation and export of glycoprotein, lipids, pectins and monomers of hemicellulose, cellulose lignin etc.

Animals—packaging and exocytosis of zymogen of exocrine pancreatic cells, mucus secretion of intestine, lactoprotein secretion by mammary gland cells, secretion of thyroxine by thyroid cells, secretion of tropocollagen and collagen, formation of melanin granules, formation of yolk and vitelline membrane of growing oocyte

3. Difference between active and passive transport

Active transport needs energy for transport of solutes against concentration gradient, where as passive transport does not need energy as transfer of molecules happens along the gradient.

Active transport happens through specific channel proteins, passive transport does not need specific channels

4.Functions of smooth endoplasmic reticulum

Synthesis of lipids

Glycogenolysis and blood glucose homeostasis

Sterol metabolism—cholesterol biosynthesis, bile acid synthesis, steroid hormone synthesis

Synthesis of triglycerides

Q.4. A) Describe <u>any one</u> of the following: **(10)**

1. The structure and chemical composition of inner mitochondrial membrane.
Ans: Description of inner mitochondrial membrane. It is multipartite with projections towards matrix side(M face), the cristae. Cristae are the site for ETS and terminal oxidation. Cristae have a head piece, a stalk and a base piece.
Chemical composition- Lipoproteinaceous, about 60% of membrane proteins. Proteins are associated with haeme, flavin,copper and non-haeme iron. Mol. Weight range 10000 to 90000 daltons. Most of the proteins are associated with 5 comlexes.(I-V).ComplesI-IV are electron transport complexes and comples V is the ATP synthesizing complex. Proportion of Sialic acid is less as compared to outer membrane.
Several ion transport components are present for transporting ions such as phosphate, ATP,ADP, dicarboxylate, tricarboxylate,gluterate, alpha keto gluterate, aspertate, pyruvate, citrulline, ornithine, bicarbonate, CO^2, Ca^{2+}, Mg^{2+}, Na^+ . Diagram with F1 particles/cristae is necessary.

2. Structure of peroxisomes. Explain their role in oxidation of compounds.
Ans: Peroxisomes are single membrane spherical organelles smaller than lysosomes. Their diameter ranges bet 0.2microm. to 1.2 microm. They contain enzymes which function in the metabolism. Their membrane is trilaminar unit membrane which is 6-8nm thick. Membrane is similar to endoplasmic membrane.
They contain oxidases and catalases. Explanation of hydrogen peroxide metabolism in peroxisomes.
Reactions are necessary.

Q. 4. B) Explain <u>any two</u> of the following: **(10)**

1. β oxidation in Glyoxysomes.
Ans: In glyoxysomes the fatty acids are oxidized to acetyl-CoA. Glyoxysomes contain enzymes that initiate the breakdown of fatty acids and additionally possess the enzymes to produce intermediate products for the synthesis of sugars by gluconeogenesis. The seedling uses these sugars synthesized from fats until it is mature enough to produce them by <u>photosynthesis</u>.
Storage lipids are initially broken down to glycerol and fatty acids in the lipid bodies. The <u>long-chain fatty acids</u> enter the glyoxysomes and are broken down to <u>acetyl-CoA</u> via the <u>β-oxidation</u> pathway. The acetyl-CoA is converted into <u>citric acid</u> from oxaloacetate and acetyl-CoA by <u>citrate</u> synthetase and then converted into isocitrate by <u>aconitase</u> in the glyoxylate pathway, which involves some of the reactions of the TCA cycle. Therefore, the glyoxysomes of higher plants are the site of β-oxidation of fatty acids and the enzymes of the <u>glyoxylate cycle</u>. Succinate is the end product of glyoxysomal metabolism of fatty acid and is not metabolized further in this <u>organelle</u>. The succinate is believed to diffuse out and it

is converted into oxaloacetate in the mitochondria. In the cytoplasm, glucose is produced by gluconeogenesis. The conversion of fat into carbohydrate is unique to glyoxysomes.
All reactions with description are necessary.

2. Anaphase in Mitosis.
Ans: Anaphase is the third phase of karyokinesis. The spindle fibres made of microtubules start shortening by loosing the units. Thus, the tension builds on centromere and the sister chomatids separate. They assume shape of letter L,V,J depending on the position of the centromere. Further shortening of fibres the daughter chromosomes reach the opposite poles.
Diagram necessary

3. Types of photosynthetic pigments in higher plants.
Ans: Photosynthetic pigments of higher plants are Chlorophull a, chlorophyll b, carotene and xanthophylls.
Their imperical formula and their role in photosynthesis should be written.

4. Structure of Thylakoid membrane.
Ans: The structure of thylakoid membrane description. Photo-system I and Photo-system II. Location of pigments in the membrane.
Diagram is necessary.

Q.5. Short notes (any 4) 20M

1. Differential centrifugation

Differential centrifugation is a common procedure in microbiology and cytology used to separate certain organelles from whole cells for further analysis of specific parts of cells. In the process, a tissue sample is first lysed to break the cell membranes and mix up the cell contents.

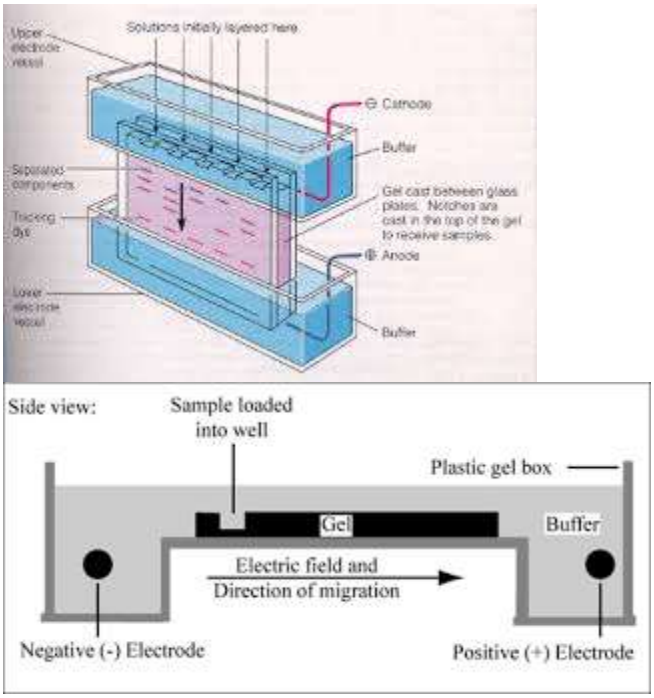
Centrifugation is the general name given to separation methods, which involves rotation about a fixed axis to produce a centrifugal (g) force. This centrifugal force forces cells down through a liquid medium and the rate of falling or sedimentation varies principally according to cell density and size. Thus, cells of different density or size sediment at different rates and at some point will be physically separate from each other. Values of sedimentation coefficients for cells are of little use as cells are rarely truly spherical and interactions among cell surfaces and the medium can occur, producing anomalous sedimentation rates. There are two basic types of centrifugation for cell separation, differential pelleting and density gradient. Density gradient centrifugation can be subdivided in two principle types, rate zonal, and isopycnic. The main difference between these two is that in isopycnic, a high-density gradient is used and cells are separated solely on differences in density. In rate zonal, a lower density gradient is used and cells are principally separated on size differences.

Differential centrifugation is a common procedure in microbiology and cytology used to separate certain organelles from whole cells for further analysis of specific parts of cells. In the process, a tissue sample is first lysed to break the cell membranes and mix up the cell contents.

Application of Differential centrifugation is a common procedure in microbiology and cytology used to separate certain organelles from whole cells for further analysis of specific

parts of cells. In the process, a tissue sample is first lysed to break the cell membranes and mix up the cell contents.

Q. 5. 2. Gel electrophoresis



Principles of DNA Gel electrophoresis

Gel electrophoresis separates **DNA** fragments by size in a solid support medium (an **agarose gel**). **DNA** samples are pipetted into the sample wells, seen as dark slots at the top of the picture. Application of an **electric current** at the top (anodal, negative) end causes the negatively-charged **DNA** [remember it's an acid] to migrate (electrophorese) towards the bottom (cathodal, positive) end. The rate of migration is proportional to size: smaller fragments move more quickly, and wind up at the bottom of the gel.

DNA is visualized by including in the gel an intercalating dye, **ethidium bromide**. **DNA** fragments take up the dye as they migrate through the gel. Illumination with **ultraviolet light** causes the intercalated dye to fluoresce with a pale pink colour.

Note that the larger fragments fluoresce more intensely. Although each of the fragments of a single class of molecule are present in **equimolar** proportions, the smaller fragments include less mass of **DNA**, take up less dye, and therefore fluoresce less intensely. This is most evident in the lane at the extreme right, which shows a "***ladder***" set of **DNA** fragments of known size that can be used to estimate the sizes of the other unknown fragments.

3. Signal hypothesis—this was proposed to explain how ribosomes which are meant for synthesis of secretory types of proteins gets specifically attached to RER membrane.

The mRNA is able to recognise free or bound ribosomes with help of special signal codons localised after the initial AUG codon. Luminal surface has signal peptidase that remove signal peptide

Diagram is expected

4. Unit membrane model

In 1935, Davson and Danielli proposed that biological membranes are made up of lipid bi-layers that are coated on both sides with thin sheets of protein and they simplified their model into the "pauci-molecular" theory. ... J. David Robertson used this method to propose the unit membrane model.According to him, the cell membrane is made of a three layer sequence of protein-lipid-protein.
The external layer is a hydrophilic layer made of protein molecules of diameter 20A−25A. The middle layer is a light hydrophobic layer made of phospholipids of diameter 25A−35A.
Diagram is must.

5. Zygotene

Ans: It is the 2nd stage of Prophase I of meiosis. The pairing of homologous chromosomes (synapsis) occurs in this phase. The pairing is zipper like, point to point. The shortening of chromosomes continues.

Diagram is necessary.

6. Chloroplast DNA

Ans: Chloroplast contains DNA. It shows cytoplasmic inheritance.

The color of the egg cell-donating branch (female parent) determined the color of the offspring.

Female parent branches that were pure green or pure white produced only pure green or pure white offspring, respectively. Female parent branches that were variegated could produce all three types of offspring, but not in any predictable ratios.

Correns speculated that some factor in the cytoplasm of the egg cell must determine the color of the offspring. It was actually a different German botanist, Erwin Baur, who suggested that the chloroplasts in the cytoplasm might carry hereditary factors (genes).

Baur thought that, in variegated plants, some of the chloroplasts must have mutations that made them unable to turn green (produce pigment).

A zygote (1-celled embryo) with mixture of chloroplasts inherited from the egg cell. Some of the chloroplasts are green, while others are white. As the zygote undergoes many rounds of mitosis to form an embryo and then a plant, the chloroplasts also divide and are distributed randomly to daughter cells at each division.

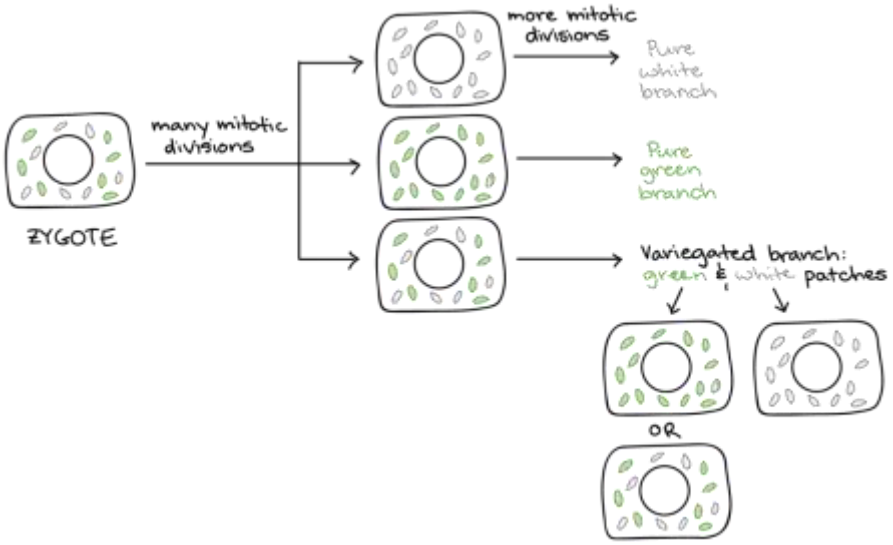


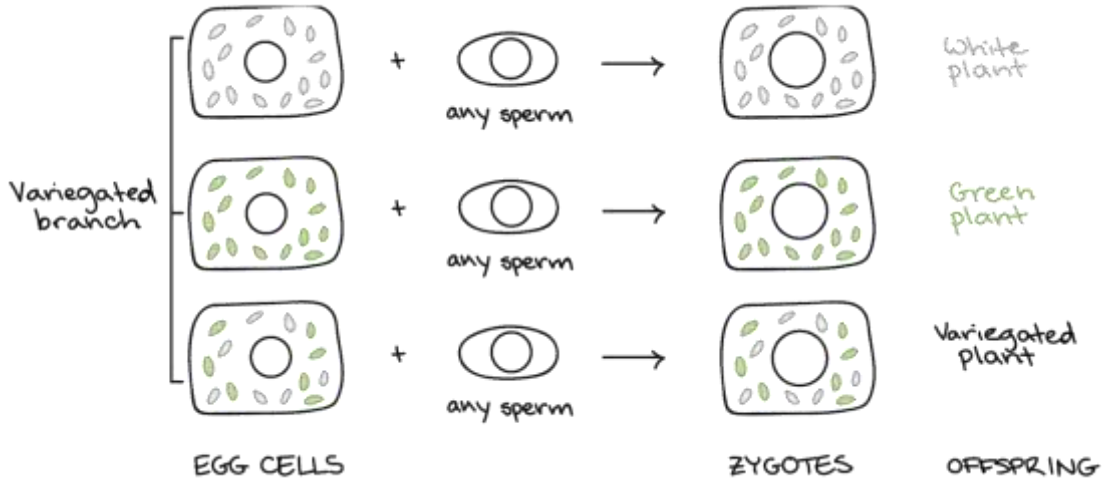
Image showing cytoplasmic segregation of chloroplasts in a plant originating from a zygote with a mixture of white (nonfunctional, mutant) chloroplasts and green (functional, normal) chloroplasts.

After many mitotic divisions in which the chloroplasts replicate and are partitioned randomly, some cells will have only green chloroplasts, others will have only white chloroplasts, and others yet will continue to have a mix. The cells with only white chloroplasts will give rise to pure white branches, and the cells with only green chloroplasts will give rise to pure green branches. The cells with a mixture of chloroplasts will give rise to variegated branches, in which ongoing random segregation of chloroplasts will produce white sectors (progeny of cells with only white chloroplasts) and green sectors (progeny of cells with mixed or green-only chloroplasts). The green cells that contain a mixture of chloroplasts will continue producing occasional pure white and pure green sectors as they divide more.

Over the many cell divisions, some cells will end up with a pure set of normal chloroplasts, making green patches). Others will get a pure set of nonfunctional chloroplasts (making white patches). Others yet will have a mix of normal and nonfunctional chloroplasts, producing green patches that may give rise to pure green or pure white sectors^77start superscript, 7, end superscript.

What about the maternal pattern of inheritance? Plants make germ cells late in development, converting cells at the tip of a branch into gamete-producing cells. A branch that’s pure green will make egg cells with green chloroplasts that give rise to pure green offspring. Similarly, a branch that's pure white will make egg cells that contain only white chloroplasts and will give rise to pure white offspring.

If a branch is variegated, it has a mixture of cells, some with only functional chloroplasts, some with only nonfunctional chloroplasts, and some with a mixture of chloroplasts. All three of these cell types may give rise to egg cells, leading to the green offspring, white offspring, and variegated offspring in unpredictable ratios.



Female branch	Egg cells	Zygotes	Offspring
Variegated branch	Egg cell with green chloroplasts, egg cell with white chloroplasts, or egg cell with mixed chloroplasts	Egg cell with white chloroplasts leads to zygote with white chloroplasts; egg cell with green chloroplasts leads to zygote with green chloroplasts; egg cell with mixture of chloroplasts leads to zygote with mixture of chloroplasts	Variegated plant