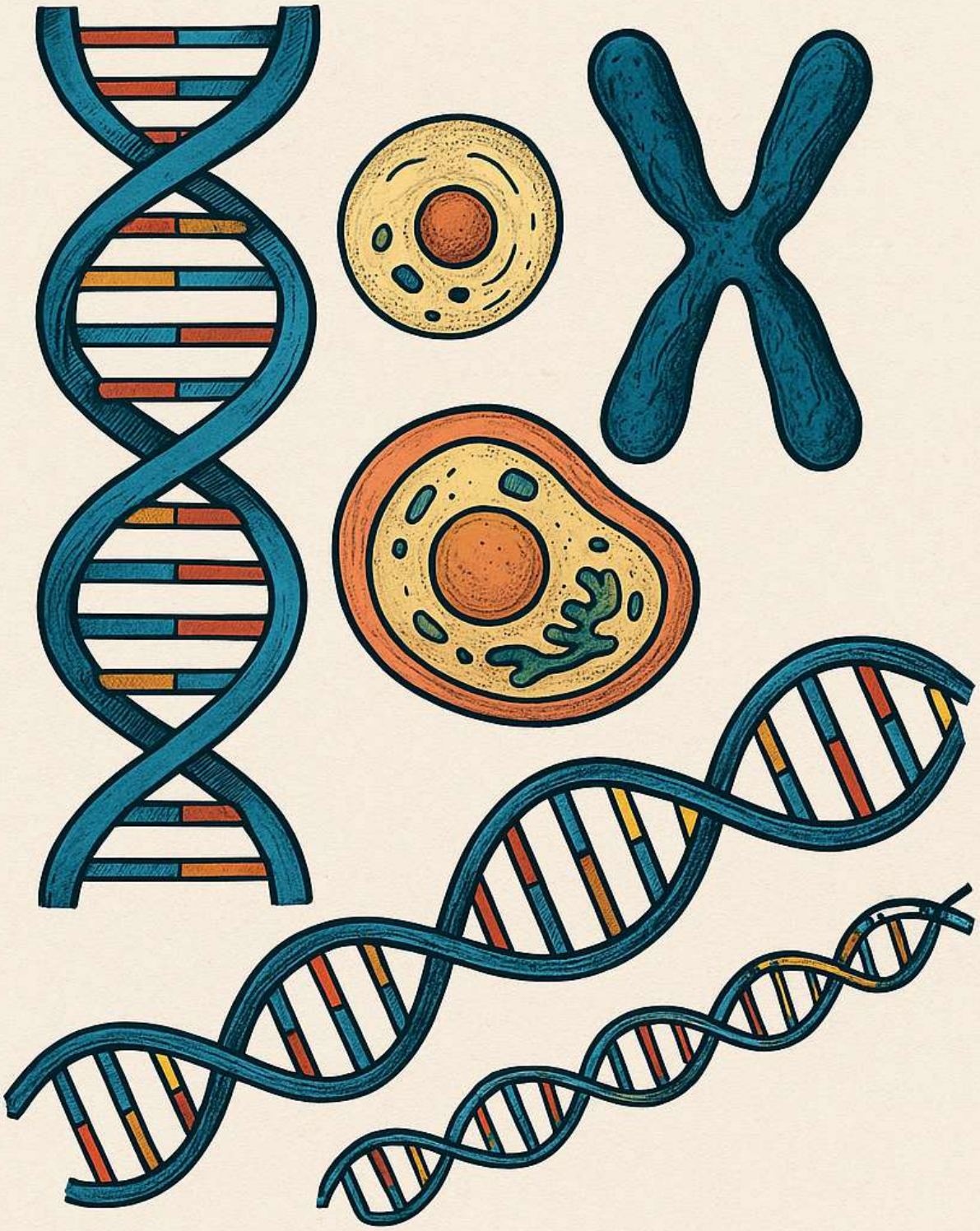
FUNDAMENTALS OF GENETICS



Fundamentals of Genetics

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CHAPTER 1

CELL AND CELL ORGANELLES

A Cell may be defined as the structural and functional unit of a living being. It is the minimal biological unit capable of maintaining and propagating itself.

A study of the structural and functional organization of different structures within a cell is known as '**Cytology**'.

Cytogenesis concerns with the study of various aspects of chromosomes and their effects on the development of characters of organisms. It is universally accepted that genes are located in chromosome. Cytogenetic originated as a result of bringing two different branches of biology namely cytology and genetics together.

History: The word „Cell" has been derived from the Latin word Cellula meaning a small compartment. The term was first used by Robert Hook (1665). Robert Hook who constructed the first compound microscope observed the sections of Cork and opined that they contain honeycomb like compartments. German biologists M.J. Schleiden and T.S. Schewann (1838) established the 'Cell theory' that all organisms are made up of cells.

One of the significant discoveries of the cell came from 'Robert Brown' (1830). He discovered the presence of a spherical body in the centre of every cell, which he named 'Nucleus'.

In 1835-37, Purkinje and Mohi independently discovered that protoplasm is an important constituent of every cell and it plays an important role in every cell activity including division.

Golgi (1838) discovered the golgi apparatus, Balbiani (181) discovered chromosomes in the salivary glands of chironomus. At amount the same time, Flemming (1882) studied cell division in detail and gave the name ' Mitosis'.

Endoplasmic reticulum was discovered by proter in 1945, while Benda gave the name mitochondria to organells originally discovered by Hemming. Lysosomes were discovered in 1955 by de Duve.

The shape of cell may be variable like spherical, rectangular, flattened, oval, polygonal triangular come like column etc.,

There is a great rang eof variation among cells in size also. This small cell size can be encountered in coccus bacteria (0.2 to 0.5 m) while the largest size of the cell is seen in Ostrich egg (Marly 15 cm).

Gross morphology of the cell

A generalized plant cell has an outer most envelope called the 'Cell wall'. This is absent in animal cells. Internal to this in the plasma membrane. This encloses the nucleus and other cytoplasmic inclusions suspended in cytoplasm. The inclusions are Ribosomes, Lysosomes, Mitochondria, Plastids, Golgi complex, Endoplasmic reticulum, Vacuole and non-liging inclusions like crystals etc.

The primitive organisms like certain bacteria blue green algae, the nucleus are not properly organized hence such cells are called Prokaryotic, while in evolved organisms, the nucleus is organized. Such cells are called Eukaryotic. The following are some of the fundamental differences between eukaryotic and prokaryotic cells.

S. No.	Character	Prokaryotic	Eukaryotic cells
1	Nuclear membrane	Absent	Present
2	DNA	Naked ad circular	Combined with proteins
3	Chromosome	Single	Multiple
4	Nucleolus	Absent	Present
5	Coty regenells	Absent	Present
6	Chromplast	Absent	Present
7	Cell wall	Non-Cellulosic	Cellulosic
8	Flagella	No definite arrangement of firbrills.	9+2 fibrillar arrangement

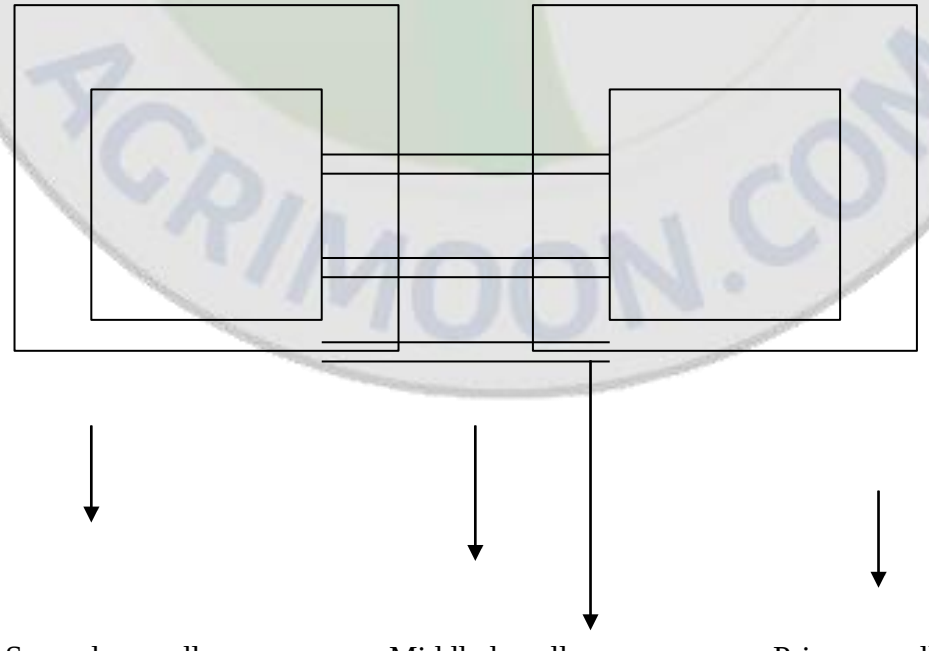
Studies with electron microscope have revealed various structures seen in an eukaryotic cell.

1. Cell wall
2. Plasma lemma
3. Endoplasmic reticular (E.R)
4. Ribosomes
5. Golgi bodies
6. Lysosomes
7. Sg herosomes
8. Chloroplasts
9. Mitochondria
10. Nucleus (Animal cell lack cell wall, chloroplast while centrioles are not found in plant cell)

1. CELL WALL: Plant cells are surrounded by a non-living and rigid coat called a 'cell wall'. The main functions of a cell wall are to provide plant cells a definite shape and mechanical support and strength to tissue and organs. Cell wall has 3 distinct parts ;

1. Middle lamella
2. Primary cell wall
3. Secondary cell wall

Middle lamella - In plants, the wall of contiguous (immediate neighbour) cells are joined by middle lamella, which is composed mainly of pectin.



Plasmodesmata

The pectin of middle lamella is most likely in the form of calcium (Ca^{++}) and Magnesium (Mg^{++}) salts. Adhesian of the walls of cotinguos cells in primarily dependant on the presence of Ca^{++} and Mg^{++} ions in themiddle lamella a removal of these ions results in the separation of cells from each other. Pectin is readily hydrolysed by the enzyme pectinase as well as by strong acids.

PRIMARY CELL WALL - is deposited after the formation of middle lamella and lies between midlle lamella and plasma lemma. It is main constituent are hemi cellulose (53%) and cellulose (30%). In addition it contains pectin (5%). Protein (5%) and lipid (7%).

SECONDARY CELL WALL - Is the last to be deposited and lies between cell wall and plasma lemma in a cell, it is the inner most laayer of wall. It is compsed of mainly of cellulose. The cellulose microfibrils are relatively more closely packed and they are arranged more or less parallel to each other. Several microfibrils associate to form a macrofibril, which is the structural unit of secondary cell wall.

2. PLASMA LEMMA (PLASMA MEMBRANE)

The membrane enclosing cytoplasm of a cell is known as plasma lemma or plasma membrane. It is composed of lipids and preoteins, the ratio between the two being quite variable among different cell types. Three distinct layers are seen under electron microscope, two or three are relatively dense and osmiphilic in nature; each of them is about 20^{o}A thich. The two osmaphilic layers are separated by a relatively light osmiphobic layer of about 35^{o}A thickness. The three layers together are known as 'Unit membrane'" this term coined by Robertson.

The chief function of plasma lemma in to regulate the movements of various molecules into and out of the cytoplasm. In addition to the passive movement of molecules, some ions are transported across plasma lemma by means of active transport.

3. CYTOPLASM

The substance, except nucelar, surrounded by the plasm lemma is known as 'Cytoplasm". Electron microscope reveal a member of membraneous and other structures in

the cytoplasm; the portion of cytoplasm other than therse structure is known as 'hyaloplasm. Of the various structures preseent in the cytoplasm, mitochondria and plastids contain DNA; as a result they are autonomous to a limited degree. However, the remaining cytoplasmic structures do not contain DNA and they are specified exclusively by nuclear genes.

The cytoplasm may contain the following structues -endoplasmic reticulum (ER), ribosomes, Golgi bodies, Lysosomes, Sphersomes, Vacoles, certioles (in animals only), microtubrils, Mitochondira and plastids (in green plants only).

4. ENDOPLASMIC RETICULUM (E.R)

The cytoplasm contains an extensive network of membrane-enclosed space; these space along with the membranes enclosing them are known as E.R. It consists of 3 types of membrane-enclosed elements.

1. Vesicles of 25-500 in μ diameter
2. Tubules of 50-100 m μ diameter
3. 40-50 m μ thick cisterns of variable length and width.

The tubulus may or may not be extensivley branched, and the cisterns may or mmay not be connected with each other.

The ultastructure of E.R membrane in the same as that of a unit membrane, that is, it has two osmiophilic layers separated by an osmiopholic layers. E.R is grouped into two categories,

1. Smooth E.R.
2. Rough E.R.

In smooth E.R elements, both outer and inner surfaces are regular and smooth. In those cells where little or no protein synthesis takes place, only smooth ER is found. The rough ER elements, ther outer surfaces of membranes have a rough appearance due to the attachment of ribiosomes on the outer surface. Rough ER is mainly composed of cisterns (membrane-enclosed plate like elements) and is found in cells actively involved in protein synthesis. Smooth and rough E.R change into each other as per the needs of cells.

Functions of ER

i. it provides the structural base for protein (rough ER), lipid, phopholipid synthesis.

ii. it proivides channel for the transport of materials synthesized in association with ER to the various parts of cells and even outside the cells.

iii. it provides a controlled passage for the export of m.RNA moelecules from nucleus to rought ER.

iv. Several enzyme molecules are embedded in the membranes of E.R.

5. RIBOSOMES

These are dense granular nucleo protein strcutures occurring in cytoplasm, matrix of mitochondria and chlorplasts. In many instances ribosomes are attached to the ER. Observed first in plants cells in 1953 by Robinson and Brown, while studying bean roots. Ranging in diameter from 150 to 200A^{o}, they have RNA and protein in equal quantities.

Ribisomes are isolated by differential centrifugation depending on sedimentation co-efficient. The sedimentation coefficient is expressed in terms of Sved berg units. The 'S' units are related with the size and weight of the ribosome molecules.

TYPES

Two types of ribosomes have been identified based on the sedimentation co-efficient. If the organella is heavier, its sedimentation co-efficient is more. The two types are 70s ribosomes and 80s ribosomes.

Ribosomes may occur singly as isolated units when they are called 'monosomes'. When they occur in clusters or groups, they are called 'polyribosomes'. The poly ribosomes may have a sedimentation coefficient of 100s-600s. The number of ribosomes per cell varies, it may be 10,000 (bacterial cell) or up to 10 million (eukaryotic cell).

Ribosomes of chlorplasts and mitochondria have their own protein synthesis. They have a sedimentation co-efficient of 55s with two sub units 40s and 30s.

ULTRA STRUCTURE

Ribosomes are oblate or spheroidal structures having two sub units (a large and a small). The larger sub unit in dome like and the smaller subunit is placed above like a cap. The 70 s ribosome has two units 50 s and 30s.

FUNCTIONS

Ribosomes are the sites of protein synthesis. The polyribosomes serve as a plat form in the assembly of amino acids brought together by specific to RNA from cytoplasm.

6. GOLGI COMPLEX

Described first by Camilo Golgi in 1890. Golgi complex found in plant cell are often referred to as 'Dictyosomes". Each golgi body consists of following parts ;

1. Eisternae
2. Tubulus
3. Vesicles
4. Golgian vacuoler.

Functions

i. Absorbtion of compounds
ii. Sites of enzyme production
iii. Sites of hormonal produciton
iv. Sits of protein storage
v. Formation of plant cell wall-by synthezing pectin, hemicellulose and cellulose microfibrils. They also help in the formation of cell plate during mitosis.

7. PLASTIDS

These are living cytoplasmic inclusions found inmost of the plants. The plastids are of three categoties viz., chromoplasts, leucoplasts and chlroplasts.

Chromoplasts

They are pigmented plastids. The pigments are non-chlorophyllous like carotenes, xeithophyll fucoxalthin, phycoerythrin etc.,

Leucoplasts

They are colourless plastids. They lack pigments and are usually present in cells which do not receives direct light. Leucoplats may be seen in the storage leaves of onion. Leucoplats that store starch are called amyloplast, those that store oil are called elaioplasts and the ones storing proteins are called alemone plasts.

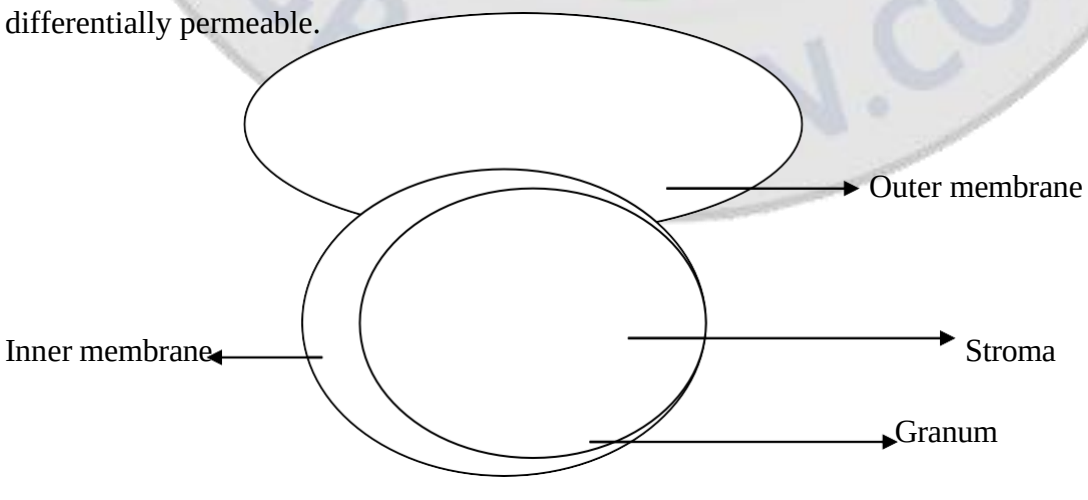
Chloroplast

These are by far, the commonest and the most plastids. As the primary sties for trapping and converting solar energy they are very vital for the existence of not only the green plants, bu for the whole living world.

Chloroplasts have varied shape and variied size. Chlorplast of polyploid cells are generally larger than in the diploid cells. They are uniformly distributed all over the cytoplasm, but in some instances they cluster towards the nucleus. The concentration of chlorplasts will also depend on light intensity.

Structure

It has a covering of two membrances with an inner membrane space. These membranes are smooth and there are no perforations or particles. The membrances are differentially permeable.



A section of chlorplasts reveals an intricate system of membranes enclosed in a granualr matrix. These membranes are called lamellae and the surrounding matrix-the stroma. In a sectional view, the lamellae can be seen packed and there stacks are called thylakoids. In higher plants, the thylakoids themselves form highly compact bundles called grana. Some thylakoids of granum extend into the stroma and maintain contact with other grama. These are called stroma thylakoid or stroma lamellae or inter grana.

Ribosomes and RNA have also been isolated from the chloroplasts indicating a mechinery for protein synthesis. Some of the important pigments present in chloroplast are chlorophylls, carotenoids, cytochromes etc.

8. NUCLEUS

It is the most important organelle of the cell which regulates all its activities. It was discovered by Robert Brown (1831). Most of the cells are uninucleate. It has the following parts;

1. Nuclear membrane
2. Karyolymph (Nuclear sap)
3. Chromonemata
4. Nucleolous
5. Endosperms

NUCLEAR MEMBRANE

It helps in effective communication between nucleus and cytoplasm. The elements of E.R. contribute to the nuclear envleope during cell division. The nuclear membrane is a double membrane with a number of pores called 'Nucleopores'. The space between these two membranes is called 'perinucelar space' or cisterna.

KARYOLYMPH (NUCLEAR SAP)

It is proteinaceous, but also has nucleic acids, enzymes and minerals. It is quite probable that in plants the nuclear sap contributes to the spindle.

CHROMONEMATA

Enclosed in the karylymph and visible in the interphase nucleus are found a number of fibrillar structures constituting a network called chromonemata or chromatrin fibrils. Some coarsegranules are deposited on the chromatin net work. These are called chromocentres and constitute the points of condensation of chromosomes. During cell division the chromatin network brakes up into specific number of chromosomes. Two regions can be identified in the chromatin material. These are heterochromatic region and enchromatic region.

The heterochromatic region stains darkly and shows numerous bead like structure called 'Chromomeres'. The heterochromatic region has less DNA. This region is believed io the genetically and metabolically inert. The light staining region of the chromatin is called the 'enchromatin region" This region contains more of DNA an is supposed to be genetically active.

NUCLEOLOUS

Nucleolous was first discovered by Fortana (1874). A speroidal body, situated either in the central or pheripheral position, the necleolous is supported to regulate the synthetic activity of the nucleus. Usually 2 or more chromosomes are associated with the nucleus (this can be seed during late prophase) and these are called nucleolar organisms as they play a role in re-appearance of the nucleolus after cell division. The number of neucleoli per nucleus varies from one to 2 or 3. Chemically the nucleolar is rich in RNA.

Functions

i. It is the active site of RNA synthesis
ii. It is the source of ribosomal RNA
iii. It produces precursors of ribosomes

ENDOSPERMS

These are granular structues present in the karyolymph and are smaller in size than the nucleolus.

CHAPTER 2

CHROMOSOME STRUCTURE AND ITS CLASSIFICATIONS

INTRODUCTION

The darkly Stained, rod shaped bodies visible under light microscope in a cell during metaphase stage of mitosis are referred to as chromosomes. Strasburger was the pioneer man who discovered chromosomes in 1875, and the term chromosome was coined by waldeyer in 1888.The main features of eukaryotic chromosomes are given below:

1. Chromosomes are not visible during interphase under light microscope. During other stages of cell division, they are visible, but are more clearly visible during mitotic metaphase. Hence, they are studied during metaphase.
2. Chromosomes bear genes in a linear fashion and thus are concerned with transmission of characters from generation to generation to generation.
3. Chromosomes of eukaryotes are enclosed by a nuclear membrane, with in prokaryotes; they remain without such envelope free in the cytoplasm.
4. Chromosomes vary in shape, size and number in different species of plants and animals.
5. Chromosomes have property of self duplication, segregation and mutation.
6. Chromosomes are composed of DNA; RNA an histones.DNA is the major genetic constituent of Chromosomes.

CHROMOSOME NUMBER

There are three types of chromosome number, vie, haploid and basic number as given below:

Haploid. It represents half of the somatic chromosome number of a species and is denoted by n since haploid chromosome number is usually found in the gametes; it is also known as game tic number.

Diploid. It refers to somatic chromosome number of a spices and is represented by 2n.Sincde diploid chromosome number is found in zygotic or somatic cells it is also referred to as zygotic or somatic number.

Basic Number. The genetic chromosome number of a true diploid species is called basic number. It is the minimum haploid chromosome number of any species which is denoted by x. For example, in wheat, the basic number is 7, where the haploid number 7,14 and 21 for diploid, tetraploid and hexaploid species, respectively. Thus haploid chromosome number differs from basic number. Both are same in case of true diploid species but differ in case of polyploid species. Thus, basic number can be a haploid number but all haploid numbers cannot be basic number. Chromosome number differs from species to species (Table 6.2). In plant kingdom, chromosome number usually is higher in dicots an pan in monocots.

Chromosome Morphology

Chromosome morphology is studied in the cell of root tip during metaphase under light microscope. Each chromosome consists of seven parts, *viz.* 1.centromere, 2. Chromatids, 3. Secondary constriction and satellite, 4. Telomeres, 5. Chromomere 6.chromonema and, 7. Matrix. A brief description of these parts is given below:

1. Centromere

The region of chromosome with witch spindle fibres are attached during metaphase is known as centromere or primary constriction or kinetochore.centromere has four important functions, viz. 1.orientation of chromosome at metaphase, 2.movement of chromosome during anaphase, 3.formation of chromatids, and 4. Chromosome shape. Since centromere is associated with movement of chromosome, viz. Terminal, sub-terminal, median, etc. Generally, each chromosome has one centromere, but in some cases, the number of centromere may vary form nil to many. Depending upon the position and number of centromere, chromosomes are given various names (Table 6.3).

2. Chromatid

One of the two distinct longitudinal subunits of a chromosome is called chromatid. These subunits of a chromosome get separated during anaphase. Chromatids are of two types‟ viz. Sister Chromatids and non sister chromatids. Sister chromatids are derived from one and the same chromosome, while non –sister chromatids originate from homologous chromosomes. Chromatids are formed due to chromosome and DNA replication during interphase. Two

chromatids of a chromosome are held together by centromere. After separation at anaphase each chromatid becomes a chromosome.

3. Secondary Constriction

The constricted or narrow region other than that of centromere is called secondary constriction. It has constant position and, therefore, can be used as useful marker. It is generally found on the short arm of a chromosome, away from the centromere. But in some cases, it is located on the long arm. A chromosome segment separated from the man body of chromosome by one secondary constriction is known as satellite. A chromosome with secondary constriction is referred to as satellite chromosome or Sat- chromosome. The Sat-chromosomes are associated with nucleolar organiser.

4. Telomere

The terminal region of a chromosome on either side is known as telomere. These are not visible in the light or electron microscopes, they are rather conceptual structures. Each chromosome has to telomeres of one chromosome cannot unit with the telomere of another chromosome due to polarity effect. In other word, translocations can occur when the ends of two chromosomes are damaged.

5. Chromomeres

The linearly arranged bead like structures found on the chromosomes is known as chromomeres. These are clearly visible in the polygene chromosome. Available evidences indicate that chromo mere represents a unite of DNA replication, chromosome coiling, RNA processing.

6. Chromonema

Under light microscope, thread like coiled structures are found in chromosomes and chromatids which are called chromonema (Plural chromonema). Chromonema is considered to be associated with three main functions. It controls size of chromosomes results in duplication of chromosomes and is the gene bearing portion of chromosomes.chromonema is a structure of sub- chromatid nature.

7. Matrix

A mass of aromatics material in which chromonemata are embedded is called matrix. Matrix is enclosed in a sheath which is knows as pellicle. Both matrix and pellicle are non genetic materials

Table: Classification and brief description of chromosome

	Basis of Classification and types of chromosomes	**Brief Description**
1.	**Position of centromere**	
.	Met centric Chromosome-	A chromosome in which centromere is located in the middle portion, such chromosomes assume V shape at anaphase.
.	Sub-meta centric - chromosome	A chromosome in which centromere is located slightly away from the canter point or has sub median position. Such chromosomes assume J shape at anaphase.
	A crocentric chromosome -	A chromosome in which centromere is located very near to one end or has sub- terminal position. Also called as sub-terminal chromosome. Such chromosome assumes J shape or rod shape during anaphase.
	Telocentric Chromosome	A chromosome in which centromere is located at one end. Such chromosome assumes rod shape during anaphase
.	Holokinetic Chromosome-	A chromosome with diffused centromere.centromere does not occupy a specific position, but is diffused throughout the body of chromosome. Whole body of such chromosome exhibits centromeric activity. Also called heliocentric chromosome.
2.	**Number of centromere**	
	A Centric chromosome	A chromosome without centromere.such chromosome remains as laggard during cell division and is eventually lost.
	Môn centric chromosome	A chromosome with one centromeres.it represents normal types of chromosome.
	Dicentirc Chromosome	A chromosome having two centromeres; such chromosomes

		make dicentric bridge at anaphase and are produced due to inversion and translocations.
3.	**Shape at anaphase**	
	V shaped Chromosome	A chromosome which assumes V shape at anaphase. It includes met centric chromosome.
	J shaped Chromosome	A chromosome which assumes J shape at anaphase. It includes sub- met centric and sub-terminal chromosome.
	Rod shaped chromosome	A chromosome with assumes rod like shape during anaphase. It includes telocentric chromosome.
4.	**Structure and appearances**	
	Linear Chromosome	A chromosome with linear structure or having both the ends free. Such chromosomes are found in eukaryotes.
	Circular chromosome	A chromosome with circular shape and structure. They are found in bacteria and viruses.
5.	**Essentiality**	
	A-Chromosome	Normal members of chromosome complements of a species which are essential for normal growth and development.
	B-Chromosome	Chromosome with are found in addition to normal chromosome complements‟ of a species. They are also celled as accessory, supernumerary or extra chromosome and are not essential for normal growth and development.
6.	**Role in Sex Determination**	
	Allosomes	Chromosome witch differ in morphology and number in male and female sex and contain sex determining genes. They are generally of two types, viz, X and Y or Z and W types (for details see under sex determination).
	Autosomes	Chromosomes which do not differ in morphology and number in male and female sex and rarely contain sex determining genes.
7.	**Structure and function**	
	Normal chromosome	Chromosome with normal structure (shape and size) and function.

	Special chromosome	Chromosomes which significantly differ and function form normal chromosome. Such chromosome include lamp brush chromosome, polytene chromosome and B – chromosome.

CHAPTER 3

CELL DIVISION

Cell is the structural and functional unit of all living organisms and all cells originates through division of pre-existing cells. Bodies of all multicellular organisms are derived from unicellular zygote through repeated divisions of zygote and the cells derived through its division. The division of chromosomes and cytoplasm of a cell into daughter cells is known as 'Cell division'. The cell that undergoes division is termed as 'parent cell', while the cells derived from the division of a parent cell are known as daughter cells.

Functions fo cell division

i. Growth and development of somatic tissue of organims
ii. Regeneration of damaged tissues
iii. Produciton of new tissues
iv. Reproduction
v. Keeping the size of cells within a limited range.

The division of pre-existing cell into daughter cells involves mainly two broad events –

a. Karyokinesis i.e. division of chromosomes of a cell into their daughter cells
b. Cytokinesis: division of cytoplasm of a cell into their daughter cells

The combined process Karyokinesis and Cytokinesis is known as cell division.

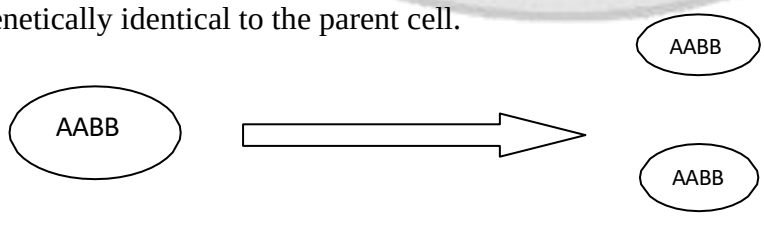
In Eukaryotic cells there are two types of cell division:

1. Mitosis and 2. Meiosis

In addition, bacterial cells divide by fission (similar to mitosis).

Mitosis:

The term mitosis was coined by Flemming in 1882. It occurs in somatic cells hence also called as somatic cell division. It involves nuclear division into two daughter nuclei each of which contain the same number and kind of chromosome in such a way that they are genetically identical to the parent cell.



Important features of mitosis:

1. It lead to production of two daughter cell from a mother cell in each cell cycle
2. The daughter cells are similar to mother cell in shape, size and chromosome complements so this is also called homotypic or equational division.
3. In plants mitosis take part in somatic organs like root tip, stem tip and leaf base which lead to growth of vegetative parts

On the basis fo chages in the morphology of necleus and the chromosomes, the events in a mitotic cell division are grouped into five stages;

i. Interphase
ii. Prophase
iii. Metaphase
iv. Anaphase
v. Telophase.

In fact, cell division is a continuous process in which a cell gradually progressed from one stage to another. So one stage merges into the next one.

INTERPHASE

In this stage of cell after the telophase of previous division and before oneset of prophase of the next one. During interphase, chromosomes are fully extented and uncoiled so that they do not takeup sufficient stain. Interphase in the longest stage. In a cell undegoing mitosis every 24 hours i.e. having a cell cycle of 24 hr., interphase may occupy upto 23 hours, while the division or mitotic phase may take up onluy 1 hour.

DNA replication occurs during the middle part of interphase. This provides the basis for classifying interphase into three substages.

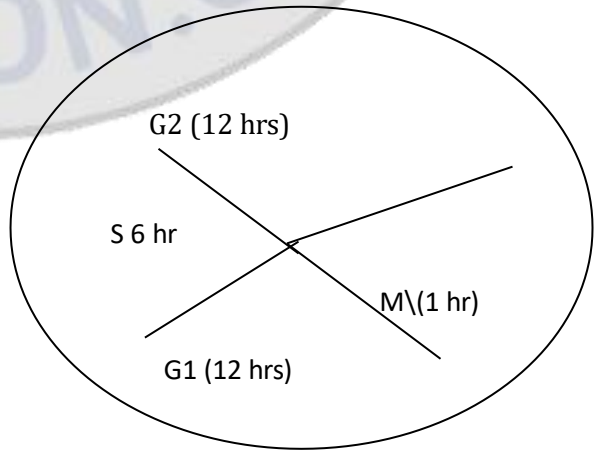
1. G1 (first gap)
2. S (Synthesis of DNA)
3. G2 (Second gap)

G1, G2 - Protein + RNA synthesis

S - DNA synthesis

M - Chromosome movement, division

Time taken in root tips fo vicia jaba.



PROPHASE

i. The appearance of define thread like strucures in nuclear is the most important event of prophase. In the beginning, chromosomes appear as a loose ball of thin wool. As prophase proceeds, chromosomes become increasingly shorter and thicker due to increased conelenstenin by mid prophase, the two chromatids of each chromosomes become visible. By the end of prophase all the chromosomes become considerably shorter and thicker.

ii. During prophase nucelolus and nuclear membrane remain present.

Chromosome condensation, that is decrease in length, with increased in thickness is mainly due to the coiling of chromosomes.

The two suiter chromatids of each chromosomes are coild inrelation to each otherl this is referred to as relational coiling. This relational coiling coiling is of two types.

i. Pleetominic coiling - The two suiter chromatids cannot separate from each other without the chromosomes being rotated. It is happening during prophase of mitosis.

ii. Paranimic coiling - Suiter chromatids are note twisted round each other, they are simply slipped into those of other they can easily separated without rotating the chromosomes. This type of coiling is found during meiotic prophase.

The relational coiling between sister chromatids goes on decreasing as the chromosomes become smaller, it disappears by late prophase.

METAPHASE

At the end of prophase four important events take place;

i. Disappearance of nucelolus

ii. Break down of nuclear envelope and distribution of its components into E.R

iii. Appearance of spindle apprartus.

iv. Arrangements of chromosomes on a single plane called 'equatorial plate'.

It is an imagigary plane and it does not represent any structural features of the cell. At meta phase, centromeres of all chromosomes lie on the equatorial plate, whiel their arms may extend outside this place.

The movement of chromosomes to an their orientation on the equatorial plate is termed metakinesis. The main features of metaphase are

i. Absenece of nucleolus

ii. Disappearance of nuclear membrane.

iii. Arrangement of chromosomes on the equatorial plate

iv. Shortest and thickest chromosomes (Condensation)

v. Coils are less in number and largest in diameter

vi. Presence of spindle appratus

vii. Absence of relation coiling between sister chromatids.

ANAPHASE

The two sister chromatids of each chromosomes separate and migrate towards the opposite poles of the cell. Anaphase begin when the centromeres of chromosomes appear to divide longitudinally so that the sister chromatids separate from each other and ends with the reachign of the chromosomes to opposite poles centromete in the first portion of each of the chromosomes to begin to move towards the poles.

Spindle fibres originate at two points located near the periphery of a cell and opposite to each other. These points are known as 'poles'

Chromosomes become somewhate more condensed as compared to those at metaphase, so that they appear relatively smaller in size.

TELOPHASE

Anaphase ends and Telaphase begins when sister chromatids of all the chromosomes of a cell reach the opposite poles. During telophase, the following events occur in the two groups of chromosomes collected at the opposite poles.

i. The chromosomes uncoil so that they become very long and thin and appeareed to be coiled into a loose ball of fine thread.

ii. Nucleus reappers

iii. Nucelar membrane is reorganised around each group of chromosomes.

iv. At the end of teophase, middle lamella appears at the equatorial plate of the cell.

The nuclear envelope dissociates into small elements which become part fo E.R. of the cell. During telophase, there elements reoriginate around the two groups of chromosomes and fuse to produce nucelar envelope around them.

In terms of duration, prophase in the longest stage of the division phase of cell cycle. In comparison anaphase is the shortest stage, while metaphase and telophase are considerably longer than anaphase.

CYTOKINESIS

It is complete by the end of Telophase. At the equatorial plate, elements of E.R. and products of golgi bodies organise and gives rise to cell plate and subsequently of cytoplasm begins in the centre of the cell and gradually extends outwards on each side in a plane, perpendicular to the axis of the spindle.

The two daughter cells produced by mitosis contain one nuclear eac; each nuclear has the same number of chromosomes as the parent cell. Each daughter cell enlarges in size till it becomes comparable to the parent cell.

MEIOSIS

Meiosis takes place during gamete formation and hence it is confined to reproductive cells only. As a consequence of meiosis, gamets contains only half (h) of the somatic chromosome number (2n). Therefore union between one male and one female gamete during fertilization restorers the chromosome number to the diploid (2n) state. Thus the chromosome number of a species remains constant from one generation to the next generation produced by sexual reproduction. In the absence of meiotic cell division, the chromosome num,ber of a species would be doubled in every generation, due to the fussion of male and female gametes, an impossible biological situation.

The nucleus of each cell undergoes two successive divisions referred to as the first and second meiotic division.

Pre-Meiotic Interphase

During 'S' phase of pre-meiotic interphase chromosomes replication takes place. But approximately 0.3% of the total DNA present in the nucleus does not replicate during the 'S' phase this DNA replicates during the zygotine substage of prophase I. A special stype of histone specific to cells preparing for meiosis is synthesized during S phase. This histone is not found in cells udnergoing mitosis, and it may be related to the entry of cells into meiosis.

FIRST MEIOTIC DIVISION

Significant events;

i. Pairing between homologous chromosomes.

ii. Crossing over between them during pachytene stage of prophase I

iii. Separation of homologus chromosomes and their migration to the opposite poles of a cell during Anaphase I. As a result, the two daughter nuclei produced by this division receive only half of the chromosomes present in somatic cells. For this reason, the first division is often referred as 'Reduction division'.

PROPHASE I : This stage is divided into 5 sub stage viz.,

i. Leptotene
ii. Zygotene
iii. Pachytene
iv. Diplotene
v. Diakinesis

LEPTOTENE

i. There is a marked increased in the nuclear volume
ii. There is chromosome condensation so that they become visibile as fine threads like a loose ball of knitting wool. Each chromosome consists of two chromatids.

ZYGOTENE

It begings with the initiation of pairing between homologous chromosomes. The main events are as follows:

i. Paring between homologous chromosomes.
ii. Completion of replication of the remaining 0.3% DNA of each nucleus, this DNA synthesis is referred to as Z-DNA synthesis or Zygote DNA synthesis.
iii. Synthesis of a specific nucelear protein
iv. Development of the synaptenemal complex and
v. Progressive condensation of chromosomes.

Pairing of homologous chromosomes is often referred as 'Synapsis'.

Synapsis

i. May begin at both ends of a homologous pair and proceed towards its centre (or)
ii. It may begin at the centromere and progress towards the telomre (or)
iii. It may begin simultaneously at several places.

PACHYTENE

It begins when synapsis comes to an end and it ends when the homologous chromosomes begin to move away from each other. The main events are ;

i. Thre is a further condensation of chromosomes, so that chromosomes pairs become shorter and thicker.

ii. Chromosomes are easily recognisable during this stage and each invalent has four chromatids.

iii. The nucleolus is distinct and quite large.

iv. Crossing over between homologous chromosomes takes place during this stage.

DIPLOTENE

i. Homologous chromosomes of ech bivalent begin to move away from each other.

ii. The two homologous of each bivalent appear to be attached with each other at one or more points, these attachments are known as chiasma. It is believed that initially chiasma are located at the points of actual crossing over between homologous chromosomes.

iii. As diplotene progress, chiasmata, slowly move towards the ends of the homologous chromosomes; this movement is referred to as chiasma terminalization i.e. movement of chaisma towards terminal positions in the chrommosomes. Chiasma terminalization occurs mainly due to the movement of homologous chromosome away from each other.

iv. There is further condensation of chromosomes so that they become progressively shorter and thicker.

DIAKINESIS

i. Bivalents move away from each other and spread towares the peripheri cells.

ii. Nucleolus, nuclear envleope disappear.

iii. The spindle appratus is organized.

The bivalents now migrate to the equatorial plate of cells; this marks the ends of diakeninsis. Bivalents may be in the form of (1) a closed ring, (2) an open ring or (3) rod shaped.

METAPHASE -I

i. Bivalents are arranged at the metaphase plate

ii. Centomeres of the two homologues of each bivalent lie on the either side of the equatorial plate.

iii. Metaphase terminates as soon as homolgous chromosomes begin to separate from each other and to migrate to opposite poles of the cell.

ANAPHASE -I

i. Separation of the two homologous chromosomes of each bivalent marks the beginning of anaphase stage.

ii. One chromosome from each bivalent begins to migrate to one pole, while the other migrates to the opposite pole.

As a result the numbner of chromosomes at each pole is exactly half (h) and each pole receives one homologue from each of the bivalents present in a cell. Thus the reduction in chromosomes number is not only a quantitative one but a qualitative one as well. Thus at the end of AI, the chromosome present is somatic cells are effectively and precisely separated into two identical groups.

TELOPHASE -I

i. The chromosomes uncoil only partially

ii. Nuclear envelope becomes organized around the two groups of chromosomes.

iii. Nucleolous also reappears.

CYTOKINENSIS

The cytoplasm of each cell divides into two halves, with a single haploid nuclear in each half. The two halves of each cell do not separate, but they staty together, and this two celled structure is known as dyad.

SECOND MEIOTIC DIVISION / MEIOSIS II

During Meiosis II, two sister chromotids of each chromosome separate and migrate to the opposite pole. As a result, the number of chromosomes in each of the two haploid nuclei remains the same (i.e haploid)., at the end of this division. The second division of meiosis is often referred to as equational division. Sometimes, it is called as 'Meiotic Mitosis'. The second meiotic division is also divided into four stages.

i. Prophase II

ii. Metaphase II

iii. Anaphase II and

iv. Telophase II

PROPHASE - II

There is no relational coiling between sister chromatids.At the end, nucelus, neclear envelope disappear and spindle apparatus is organised.

Cytokinesis

Dyad divides into two parts. One parent cell produces from haploid daughter cells after meiosis. The four daughter cells present together and are known as tetrad.

CHAPTER 4

MENDEL'S LAWS OF INHERITANCE

Introduction:

Mendel was borne in 1822 near Brunn (Czechoslovakia) in Austria, in the family of a poor farmer. Unable to continue his studies, due to poverty, he joined St. Augustinian Monastry at Brunn in 1843 and became a priest. He was sent to the University of Vienna, where he studied physics, maths and philosophy etc., Then he returned to Brunn in 1854 where he was appointed as a substitute science teacher and his peformance as a science teacher was excellent. In addition he worked as a priest in the local church. He lived in a house located within the premises of the church. He began to collect pea seeds for his experiments in 1857 from commercial seed growers all over the Europe. He conducted all his experiments within the kitchen garden of his house with the help of his own resources.

After 7 years, he presented his findings byne the Natural History Society of Brunn in 1865. This paper entitled " Experiments in plant hybridization" was presented in German language. Later Mendel studied on Honey bee, some other plants and climatology. He died in 1884 at an age of 62 years and long before the world understood and appreaciated his contributions to our understanding of life.

Sixteen years after his demise, three scientists working independently of each other de vries in Hollad, correns in Germany and Tschermak in Austria, arrived at the same conclusions as those of Mendel. After this rediscovery there was a spirit of interest in the Mendel's findings and the science of genetics was timely borne. Although the basic principles of genetics wre enuciated in 1865 itself, the new baby borne was kept in an incubator and forgotten for the next 35 years.

PEA as an experimental material

Pea offered several advantages as an experiment material.

i. In the pea varieties available commercially, several characters had two contrasting form which were easily distinguishable from each other.

Character	Dominent form	Recessive form
Seed shape	Round	Wrinkled

Seed coat colour	Grey	White
Cotyledon colour	Yellow	Green
Pod colour	Green	Yellow
Pod shaped	Full	Constricted
Position of flowers	Axial	Terminal
Length of stem	Tall	Dwarf

ii. The flower strcuture of pear ensured self pollination this was experimentally verified by Mendel. This greatly facilitated the production of F2 and F3 progeny as well as avoided contamination by foreign pollen.

iii. Pea flowers are relatively large. Therefore emasculation and pollination is quite easy, which allows easy artificial hybridization in pea.

iv. The duration of pea crop is of a single season. As a result, every year one generation of pea can be grown.

v. Pea seeds are large and present no problem in germination. Pea plants are relatively easy to grow and each plant occupies only a small space. This persists a large number of plants to be grown in a relatively small area.
(In addition, Mendel worked in Raj mash, *P. vulgaris*)

Reason for Mendel's success

i. Mendel studied the inheritance of only one pair of contrastiang chracters at a time. This allowed him to classify in F2, F2 progenies into two clear cut groups.

ii. He selected pea varieties that had clearly different gorms of one or more chracters.

iii. Mendel classified all the plants of a population on the basin of the contrasting chracters under study and kept an accurate record of the numbner of plants in each category.

iv. Mendel carried out his experiments with great care and elaborators. For e.g. He grew the pea varieties used as parents for two seasons to avoid mechanical mixtures and the verify homozygosity of varieties and stability of the character difference.

v. His knowledge of maths was a definite asset on interpretation of his findings.
e.g. He was able accept the ratios ranging from 2.82:1 to 3.15:1 over all estimation of 3:1 and not separate ratio.

vi. Mendel was able to formulate appropriate hypothesis on the basis of explanation he offered for his experimental findings. Further, he proceeded to test these hypothesis experimentally to prove the correctness of his explanations.

MENDEL WAS UNDOUBTEDLY LUCKY

i. Seven characters selected by Mendel showed qualitative inheritance.

ii. Each characters is governed by a single dominant gene.

iii. Of the 7 characters, the gene for 2 character were located in one chromosome. While 3 other were in another chromosome. But out the these, only 2 were close enough to distort di hybrid ratio of 9:3:3:1. Luckily Mendel did not study this character pair.

REASONS FOR NEGLECT OF MENDEL'S FINDINGS

i. Mendel used mathematical principles of probability to explain a biological phenomena. This was something new and not radily acceptable to biologist.

ii. He studied constrasting pairs of characters exhibiting discontinueous variation, which is unimportant in evolution.

iii. In this studies, only the parental forms appeared, no new forms (variation) were recovered.

iv. The pheneomenon of fertilization, behaviour of chromosomes during cell division wre not known at the time, when Mendel presented his findings.

v. Mendel failed to demonstrate his conclusion in other species.

General Terminology:

(1) Hereditary factor: The entities which are responsible for transmission of characters from one generation to next generation are called hereditary factor.

(2) Contrasting characters: The individual plants with marked phenotypic difference are known as contrasting characters.

(3) Dominant and recessive characters: When a cross is made between plants having contrasting characters, only one character is expressed in the hybrid and the other is suppressed. The characters which is expressed is called dominant character and the character which is suppressed is known as recessive characters.

(4) Homozygous condition: The individuals having similar alleles on the corresponding locus of homologous chromosome.

(5) Heterozygous condition: The individuals having dissimilar alleles on the corresponding locus of homologous chromosome.

(6) Phenotypes: It refers to the morphological features of an individual‟s like color, height, sex etc.

(7) Genotypes: It refers to the genetic constitution of an individual‟s like AA, Aa and aa.

(8) Monohybrid Cross: The cross involving one pair affecting one character. Like RR (Red), rr (White) and Like Cross for Flower colour

RR X rr

Red White

Rr

Gametes	**R**	**r**
R	RR	Rr
r	Rr	rr

Genotypic ratio: 1 RR: 2 Rr: 1rr

Phenotypic ratio: 3 Red: 1 White

(9) Dihybrid Cross: The cross involving two gene pair and each gene pair affecting different character. Like Tall and Yellow plant crossed with Dwarf and white plant

TTYY X ttyy

Tall and Yellow Dwarf and white

TtYy

Gametes	**TY**	**Ty**	**tY**	**ty**
TY	TTYY	TTYy	TtYY	TtYy
Ty	TTYy	TTyy	TtYy	Ttyy

tY	TtYY	TtYy	ttYY	ttYy
Ty	TtYy	Ttyy	ttYy	ttyy

Genotypic ratio: 1 TTYY: 2TTYy: 2TtYY: 4TtYy: 1TTyy: 2Ttyy: 1ttYY: 2ttYy: 1ttyy
Phenotypic ratio: 9 Tall and Yellow: 3 Tall and White: 3 Dwarf and White: 1 Dwarf and White

Backcross and testcross

Backcross in a cross between hybrid and any one of the parents, whereas testcross is a cross between hybrid and a recessive homozygote.

F1 Test cross Back cross

Rr x rr Rr x RR or rr

R r r

Rr : rr

1 Round : 1 Wrinkled

LAWS OF MENDEL

MENDEL'S FIRST LAW (LAW OF SEGREGATION)

When a pair of contrastiang characters are brought together in a hybrid, the factors responsible for the character do not contaminate each other in the hybrid, but when gametes are formed they segregate and pass into different gametes in a difinite proportion.

In fertilization, the gametes combine at random (i.e they unite freely in all possible combinations). The F2 consists of 4 combinations viz., RR, Rr, rR, rr in equal numbers.

Law of Segragation: The two alleles of a gene remain separate and do not contaminate blend or each other in the F_1 or hybrid. At the time of gamete formation in F_1, the two alleles separate and pass into different gametes. E.g

RR X rr

Red White

F_1 Rr

Gamets	R	r
R	RR Red	Rr Red
R	Rr Red	Rr White

Genotypic ratio: 1 RR: 2 Rr: 1rr

Phenotypic ratio: 3 Red: 1 White

There is no visible indication of the presence of allele 'r' in the F1, the allele R and r do not linked or fuse with each other while they are together in F1. The alleles R and r do not also contaminate or affect each other.

In F1, all the offspring were uniform and resembled one of the parents so closely that the characters of the other escaped observation completely. Those parental chracters which aappeard in F1 were termed dominant, and those parental characters which entirely disappeared in F1 were termed 'Recessive'.

Exceptions of Mendel's laws of Segregation

Incomplete Dominance: When the intensity of phenotype produced by heterozygotes is less than that produced by homozygotes for the concern dominant allele.

The phenotype of heterozygotes falls between those of the homozygotes for the homozygotes for the concerned alleles, such situation is known as incomplete or partial dominance.

E.g. Flower colour in Four „o‟ clock (Mirabilis Jalapa)

RR X rr

Red ⇓ White

Rr

Pink

⇓

Gamets	R	r
R	RR	Rr
R	Rr	rr

Genotypic ratio: 1 RR: 2 Rr: 1rr

Phenotypic ratio: 1 Red: 2 Pink: 1 White

Codominant: When the heterozygotes for the genes possess the phenotype produced by the expression of both the concern alleles, such situation is known as Codominance.

E.g. ABO blood group in human being:

AA X BB

⇓

AB

⇓

Gametes	A	B
A	AA	AB
B	AB	BB

Genotypic ratio: 1 AA: 2 AB: 1BB

Phenotypic ratio: 1 A Type: 2 AB type: 1 B Type

Over dominant: In case of some genes, the intensity of a characters expression is greater in the heterozygotes than in the two concerned homozygotes. Such situation is known as overdominance

E.g. Sepiteridin eye pigment in Drosophilla

WW X ww

Normal dull red eye colour ⇩ White eye

F_1 generation Ww

Dark red eye colour

⇩

Gametes	W	w
W	WW	Ww
w	Ww	ww

F_2 generation :

Genotypic ratio: 1 WW: 2 Ww: 1ww

Phenotypic ratio: 1 Normal dull red eye colour: 2 Dark red eye colour type: 1 White eye Type

Reciprocal crosses

It is a second cross involving the same characters as the first but with the sexes of the parents interchanged.

Whichever way the cross is made, the results will be the same, in case nuclear genes determine the characters. However, when heriditory factors, in the cytoplasm also interact with nuclear genes, receiprocal differences have been observed. In representing crosses, it is conventional to write the female parent first and the male parent second.

Xenia

Effect of pollen on the embryo and endosperm. e.g. in maize, colourless seeded plant is dusted with purple seeded plant pollen, shows the purple seed in the cob.

Purple is dominant over colourless.

Law of Independent Assortment: The segregation of two or more characters in the same hybrid is independent of each other. When an individual forms gametes, the member of a pair of alleles always segregate frome ach other but the members of different pair of alleles assort independent of each other.

TTYY X ttyy

Tall and Yellow ⇩ Dwarf and white

F_1 TtYy

⇩

F_2 population produced after selfing of F_1

Gametes	**TY**	**Ty**	**tY**	**ty**
TY	TTYY	TTYy	TtYY	TtYy
Ty	TTYy	TTyy	TtYy	Ttyy
tY	TtYY	TtYy	ttYY	ttYy
ty	TtYy	Ttyy	ttYy	ttyy

Genotypic and Phenotypic ratio:

Types of genotypes	Genotypic ratio	Phenotype	General Genotype	Phenotype ratio
TTYY	1	Tall and Yellow	T_Y_	9
TTYy	2	Tall and Yellow		
TtYY	2	Tall and Yellow		
TtYy	4	Tall and Yellow		
TTyy	1	Tall and White	T_yy	3
Ttyy	2	Tall and White		
ttYY	1	Dwarf and White	ttY_	3
ttYy	2	Dwarf and White		
ttyy	1	Dwarf and White	ttyy	1

CHAPTER 5
EPISTASIS AND GENE INTERACTION

When an individual forms gametes, the two members of each pair of alleles always separate from each other but the separation in one pair of alleles is independent of the separation in any other pair of alleles. Gametes, therefore, always contain any one allele of each of the several pairs of alleles found in an individual. At fertilisation, these gametes recombine at random to give rise to new individuals.

When different pairs of alleles influence the same character of an individual, it is likely that the expressions of these genes interact. As two different genes interact and affect the same character, such a genetic interaction is said to be intergenic or nonallelic. In nonallelic interactions different genes located on the same or different chromosomes interact with one another for the expression of a single phenotypic trait of an organism.

Intergenic or nonallelic interactions may suppress or mask the action of a gene at another locus or modify partially or completely the effect of another gene. This nonallelic interaction is otherwise called *epistasis.*

Definition: - A kind of interaction between genes belonging to different pairs of alleles, the dominant allele in one of the pairs preventing the dominant allele in the other pair from expressing itself. Thus, the gene A may be epistatic over B. B is then said to be hypostatic to A.

We shall now consider a few cases of independently transmitted pairs of alleles that are not independent in their expression.

Intergenic Non-epistatic interaction (9: 3 : 3 : 1 Ratio)

A classical case of two pairs of alleles affecting the same characteristic and producing in the F_2 four different phenotypes in the ratio of 9: 3: 3: 1 was discovered in fowls by Bateson and Punnett.

Each breed of pultry possesses a characteristic type of comb. The Wyandotte breed has a comb known as the „rose" comb, the Brahma has a „pea" comb, the Leghorn has a „single" comb and the Malay breed has a comb known as the „walnut" comb. Each of these breeds true.

Crosses between rose-combed and single-combed types show that rose is dominant to single comb and that there is a segregation of 3 rose: 1 single comb in the F_2. In matings between pea-combed and single-combed birds, pea comb is found to be dominant over single comb and a 3 : 1 ratio appears in the F_2.

When, however, a rose-combed fowl is crossed with a pea combed one, all the F_1 birds show the walnut comb. When the F_1 walnut combed birds are bred together, there appears in the F_2 9 walnut : 3 rose : 3 pea : 1 single comb.

These results can be interpreted as follows: The rose comb is due to a gene R and the pea is due to another gene P. The walnut comb is due to the presence of both the dominant genes, R and P and the single comb is due to their recessive alleles, r and p.

The breeding behaviour of the different genotypes of the F_2 is summarised. F_2

Phenotype	Genotype	Ratio	Breeding behaviour
Walnut	RRPP	1	All the progeny walnut-combed
	RRPp	2	3 walnut (RP) : 1 rose (Rp)
	RrPP	2	3 walnut (RP) : 1 pea (rP)
	RrPp	4	9 walnut : 3 rose : 3 pea : 1 single
Rose	RRpp	1	All the progeny rose-combed
	Rrpp	2	3 rose (Rp) : 1 single (rp)
Pea	rrPP	1	All the progeny pea-combed
	rrPp	2	3 pea (rP) : 1 single (rp)
Single	rrpp	1	All the progeny single-combed

The above example depicts a case of non-epistatic intergenic interaction in which two genes that determine the same character produce a new phenotype by mutual non-epistatic interaction.

Difference between dominance and epistasis

The phenomenon of dominance involve intra-genic or inter-allelic gene suppression, or the masking effect which one allele has upon the expression of another allele at the same locus, while the phenomenon of epistasis involves inter-genic suppression or the masking effect which one gene locus has upon the expression of another. The classical phenotypic ratio of

9:3:3:1 observed in the progeny of dihybrid parents becomes modified by epistasis into ratios which are various combinations of the 9:3:3:1 groupings.

Types of epistasis

1. Dominant epistasis (12:3:1),
2. Recessive epistasis (9:3:4)
3. Duplicate dominant epistasis (15:1)
4. Duplicate recessive epistasis (9:7)
5. Dominant and recessive epistasis (13:3)
6. Duplicate genes with cumulatie effect (9: 6 : 1)

1. Dominant Epistasis (12 : 3 : 1 Ratio)

In *Sorghum*, pearly grains are shining, translucent and oily white, and chalky grains are not shining but opaque and dull white. When a plant with pearly grains and another with chalky grains are crossed the F_1 is pearly. In the F_2 there is a segregation into 3 pearly : 1 chalky. The gene for pearly grains can be represented by Z and the gene for chalky by z.

The colour of the grains may be either red or white. When a plant with red grains is crossed with one with white grains, the F_1 is red and the F_2 shows a segregation of 3 red : 1 white. The gene for red grains is represented by W and white by w.

When the colour of the grain is white, it is possible to say whether it is pearly or chalky, but when the colour is red, it is not possible to find out whether it is pearly or chalky. One character, the red colour of the grain, masks another character, the pearliness

When two non-allelic genes affect the same part or trait of an organism, it is likely that the expression of one covers up or hides the expression of the other. A gene which thus masks the expression of another gene which is not its allele is said to be **epistatic** to it and the gene which is hidden is said to be **hypostatic**. Epistasis is the dominating influence of one gene over another which is not its allele and is similar to dominance expect that it occurs between different genes instead of between the members of an allelic pair. Dominant epistasis is also called Dominant suppressor.

The gene W is epistatic to those for pearliness Z and chalkiness z and so long as it is present, pearlines or chalkiness cannot be distinguished from one another.

Where this gene W is lacking, i.e., where the genotype is ww, the grains will be pearly if Z is present and chalky if Z is absent.

Thus W masks everything that is hypostatic to it so that Z which segregates quite independently of W produces a visible expression only when W is absent.

	3Z	=	9 WZ	Red
3W	1Z	=	3WZ	
	3Z	=	3 wZ	White pearly
1w				
	1Z	=	1wz	White chalky

The F_2 thus shows a ratio of 12 red : 3 white pearly : 1 white chalky.

2. Recessive Epistasis (9 : 3 : 4 Ratio)

In a cross between a cholam (*Sorghum)* plant with blackish purple leaf sheath and another with brown leaf sheath, the F_1 is found to be blackish purple. In the F_2 there is segregation into 3 blackish purple : 1 brown. The gene for blackish purple can therefore be represented by P and brown by p.

In another cross between a blackish purple and a brown plant, the F_1 is found to be reddish purple. The F_1 is expected to be blackish purple because blackish purple is dominant to brown. Actually however, the F_1 is reddish purple. Evidently, there is another gene which is responsible for converting the blackish purple colour into reddish purple. This gene is called a supplementary gene, Q and it adds to the effects of blackish purple.

In the F_2 there is a segregation into 9 reddish purple : 3 blackish purple : 4 brown.

The gene P is responsible for the blackish purple colour and its allele p for the brown colour. When the supplementary gene Q is found in combination with the gene P for blackish purple, the leaf sheath is reddish purple. The gene Q, however, has no phenotypic expression of its own and plants will therefore be brown whether they possess Q or not, if they lack the gene P.

Blackish purple		X	Brown
PPqq			**ppQQ**
F_1 Reddish purple		**PpQq**	

Genotypic ratio in the F_2

	1 **QQ**	= 1 **PPQQ**	Reddish purple
1 **PP**	2 **Qq**	= 2 **PPQq**	Reddish purple
	1 **qq**	= 1 **PPqq**	Blackish purple

	1 **QQ**	= 2 **PpQQ**	Reddish purple
2 **Pp**	2 **Qq**	= 4 **PpQq**	Reddish purple
	1 **qq**	= 2 **Ppqq**	Blackish purple
	1 **QQ**	= 1 **ppQQ**	Brown
1 **pp**	2 **Qq**	= 2 **ppQq**	Brown
	1 **qq**	= 1 **ppqq**	Brown

Phenotypic ratio in the F_2

	3 **Q**	= 9 **PQ**	Reddish purple
3 **P**	1 **q**	= 3 **Pq**	Blackish purple
1p	3 **Q**	= 3 **pQ**	Brown
	1q	= 1 **pq**	

This type of epistasis is called recessive epistasis, since the recessive allele of one gene masks the phenotypic expression of the dominant or recessive allele of another gene.

Duplicate dominant epistasis (15 : 1)

In jowar (Sorghum), plants with starchy grains breed true.

In a cross between a plant with starchy grains and a plant with waxy grains, the F_1 is starchy and the F_2 shows a segregation of 3 starchy : 1 waxy.

In a cross between a second plant with starchy grains and a plant with waxy grains, the F_1 is again found to be starchy and the F2 again shows a segregation of 3 starchy : 1 waxy.

In a cross between the first plant with starchy grains and the second plant with starchy grains, the F_1 is found to be starchy but the F_2 shows a segregation of 15 starchy : 1 waxy.

Starchy grain is evidently due to the presence of a dominant gene Wx, (for the sake of simplicity, denoted as W_1) or another dominant gene Wx_2 (for the sake of simplicity, denoted as W_2) or both. When both the dominant genes are absent, the grain is waxy.

Duplicate genes are two pairs of alleles, either alone or together, producing the same effect. They are identical genes but are situated on two different pairs of chromosome. Each gene is dominate to its allele but does not add to the effect of the other. It is conventional to designate two such genes by the same letter followed by different numerical subscripts, as W_1 andW_2.

Duplicate recessive epistasis (9:7)

In *Sorghum*, plants with white grains, when self fertilised, produce progeny all of which have white grains, i.e., they breed true. When however, two true-breeding white grained plants are artificially crossed, the F1 is not white but brown. In the F_2, brown and white appear in the proportion of 9 7.

Since the white-grained plants are pure-breeding, they are homozygous but they cannot have identical genotypes, because the two white-grained parents, when crossed, give rise to brown grained progeny. The two white-grained parents are therefore homozygous but with different genotypes

The brown colour of the grains is presumably due to the presence of two dominant genes, B_1 and B_2. The two white-grained parents are therefore homozygous for one or other of these two dominant genes but because they have different genotypes (due to the fact that they give rise to brown-grained progeny on hybridisation) one white-grained parent is assumed to be $B_1B_1b_2b_2$ and the other white-grained parent to be $b_1b_1B_2B_2$. One parent is white because it lacks B_2 and the other parent is white because it lacks B_1 but a cross between two such white-grained parents produces offspring with both the dominant genes B_1 and B_2 and consequently, brown grains. These genes responsible for the brown colour of grains are called complementary genes.

Two non-allelic dominant genes that act together to produce a phenotype different from that produced by the homozygous recessive of the one or the other or both are called complementary genes.

Complementary genes are differentiated from one another by addicting different alphabetic subscripts to the same letter, e.g., B_m and B_n but, for the sake of simplicity, the symbols B1 and B2 have been adopted in the present discussion.

The expectations in the F_2 from a cross between two plants with white grains are shown.

The expectations in the F_2 from a cross between white-grained Sorghum plants

Complementary genes in sweet peas

Bateson and Punnett discovered that the F_1 of a cross between two white flowered strains of the Emily Henderson sweet pea, Lathyrus odoratus, was purple flowered. When the F_1 plants were self-fertilised, they produced offspring consisting of about nine-sixteenths purple flowered plants and seven-sixteenths white flowered ones.

Both parents were found to be true breeding and therefore homozygous. The were phenotypically identical in every respect but genotypically different, as otherwise, the F1 would have been white flowered.

It was therefore conclude that the purple colour results from an interaction between two different dominant genes, on e from one white flowered parent and the other from the other white flowered parent and the white colour is due to the absence of either or both of these dominant genes.

P White X White

$P_1P_1p_2p_2$ $p_1p_1P_2P_2$

F_1 Purple

$P_1p_1P_2p_2$

F_2 $9\ P_1 - P_2 -$ = 9 purple

$3\ P_1 - p_2p_2$

$3\ p_1p_1P_2 -$ = 7 White

$1p_1p_1P_2P_2$

Dominant and Recessive epistasis (13 : 3)

In crosses between Sorghum plants with purple node and plants with green node, the F_1 hybrids are with purple node. In the F_2 there is a segregation into 3 purple : 1 green. The gene for purple node P_j (denoted for convenience as P) is therefore dominant over that for green p (denoted as p).

In a certain cross between a Sorghum plant with purple node and one with green node, the F1 is with green node. Since purple is dominant over green, the F_1 is excepted to be purple but it is observed to be green. The gene for purple node is unable to express itself probably because of the presence of another gene. This gene is called an inhibitory gene and is represented by I. this gene is capable of inhibiting the production of purple colour in plants with P.

Among the F_2, 13 are green and 3 are purple. This is because plants are purple only if they possess the gene for purple colour P in the absence of the inhibitory gene. In the presence of the inhibitory gene I, plants with the gene for purple P are unable to exhibit the purple colour and are only green. Plants which do not have the gene for purple colour are green whether they have the inhibitory gene or not.

The inhibitory gene I has thus no phenotypic expression of its own and its presence can be judged only by its effect on the gene for purple P.

Duplicate genes with cumulative effect (9 : 6 : 1)

In barley, plants with light purple grains breed true.

In a cross between a plant with light purple grains and a plant with white grains, the F_1 is light purple and the F_2 shows a segregation of 3 light purple : 1 white.

In a cross between a second plant with light purple grains and a plant with white grains, the F_1 is light purple and the F_2 again shows a segregation of 3 light purple : 1 white.

In cross between a first plant with light purple grains and the second plant with light purple grins, the F_1 is found to be dark purple. The F_2 segregation is 9 dark purple : 6 light purple : 1 white.

Light purple colour of the grain is evidently due to the presence of a dominant gene P_1 or another dominant gene P_2. The two non-alleliuc dominant genes P_1 and P_2 possess an additive effect and the colour of the grain is dark purple when the gene P_1 and P_2 are present together.

When both the dominant genes are absent, the colour of the grain in white.

P Light purple x Light purple

$\mathbf{P_1P_1p_2p_2}$ $\mathbf{p_1p_1P_2P_2}$

F_1 Dark purple

$\mathbf{P_1p_1P_2p_2}$

Genotypic ratio in the F_2

	$1\ \mathbf{P_2P_2}$	$= 2\ \mathbf{P_1p_1P_2P_2}$ Dark
$1\ \mathbf{P_1P_1}$	$2\ \mathbf{P_2p_2}$	$= 2\ \mathbf{P_1P_1P_2p_2}$ Dark
	$1\ \mathbf{p_2p_2}$	$= 1\ \mathbf{P_1P_1p_2p_2}$ Light
	$1\ \mathbf{P_2P_2}$	$= 2\ \mathbf{P_1p_1P_2P_2}$ Dark
$2\ \mathbf{P_1p_1}$	$2\ \mathbf{P_2p_2}$	$= 4\ \mathbf{P_1p_1P_2p_2}$ Dark
	$1\ \mathbf{p_2p_2}$	$= 2\ \mathbf{P_1p_1p_2p_2}$ Light
	$1\ \mathbf{P_2P_2}$	$= 1\ \mathbf{p_1p_1P_2P_2}$ Dark

1 $p_1 p_1$ 2 $P_2 p_2$ = 2 $p_1 p_1 P_2 p_2$ Dark

1$p_2 p_2$=1$p_1 p_1 p_2 p_2$ Light

The inheritance of grain colour in wheat has been shown by Nilsson – Ehle to be similar to that in barley except that, in wheat, the genes are incompletely dominant over their alleles. Thus the additive or cumulative action of the dominant alleles of two non-allelic genes causes the full expression of a phenotype, as distinctly different from the presence of the dominant allele of any one of the genes

PLEIOTROPISM

A single gene may sometimes affect more than one chracteristics fo the organism eg. In cotton, Punjab hairy lintless gene ' lic'. It produces;

i. Seeds which are without lint.
ii. In complete laciniation of the bay
iii. Reduction in the number and length of internodes
iv. Reduction in boll size and fertility.

When a gene courses changes in two or more parts or characters that are not obviously related, the gene is called 'pleiotropic gene'.

Multiple or marigold phenotypic expression of a single gene is called 'pleiortropism'.

MULTIPLE ALLELES

The alternative form of a gene is called allele. Generally it was assumed that a single gene has two alternative forms, one of them is dominant over recessive allele. But in many other cases several alleles of a single gene are known, each governing a distinct form of the concerned trait. Such situation is known as multiple allelism and the many alleles of a single gene are called multiple alleles.

Characteristics:

1. A gene can have more than two alleles, but a diploid individual only has one or two of them.

2. Different allele combinations can produce different phenotypes and different severities of symptoms.

Features

1. Multiple alleles are always at the same locus in the homologus chromosome.
2. There is no crossing over with in a multiple allelic series. When two alleles are involved in a crosss, the same two alleles are recovered in the F12 or test cross progeny.
3. Multiple alleles always affect the same characters.

4. The wild type allele is naturally always dominant.

E.g. 1 Colour corolla in Asiatic cotton

Full yellow ⟶ YY, YY^{p} < Yy

Pale ⟶ Y^{p} Y^{p}, Y^{p}y

White ⟶ yy

Degree of dominance Y > Y^{p} > y

Eg.2 Fur colou in Rabbit is governed by four allele of a single gene c. each allele has different phenotype like c (albino/ no pigment/ white fur colour), c^{h} (Himalayan/white body and tip of ear, feet, face and tail are black), c^{ch} (Chinchilla/ mixture of white and coloured hairs over the whole body), C or c^{+} (Full body coloured)

Dominant relationship: C or c^{+} > c^{ch} > c^{h} > c

Allele	Genotypes	phenotypes
c	Cc	albino/ no pigment/ white fur colour
c^{h}	c^{h} c^{h}, c^{h}c	Himalayan/white body and tip of ear, feet, face and tail are black
c^{ch}	c^{ch} c^{ch}, c^{ch} c^{h}, c^{ch} c	Chinchilla/ mixture of white and coloured hairs over the whole body
C or c^{+}	c^{+} c^{+}, c^{+} c^{ch}, c^{+} c^{h} ,c^{+} c	Full body coloured

Eg.3. The ABO blood type system is also an example of a trait that is controlled by more than just a single pair of alleles. In other words, it is due to a multiple-allele series. In this case, there are three alleles (A, B, and O), but each individual only inherits two of them (one from each parent).

eg. 3. A-B-O blood group in human beings three allels I^{A} . I^{B}, I^{O}, Where I^{A} and I^{B} are codomiant. (I^{A} = and I^{B}) > I^{o}

Genotype	**Phenotype**
I^{A} I^{A}, I^{A} I^{O}	'A' group
I^{B} I^{B}, I^{B} I^{0}	'B' group
I^{A} I^{B}	AB group
I^{o} I^{o}	'O' group.

PSEUDO ALLELES

Non allele so closely linked as often inherited as one gone, but are separate from each other. (by cross over studies).

These effects are found in Drosophila, corn, cotton, bacteria, Vrisues.

ISO ALLELES

Usually wild type alleles (respresented as +) is dominant over its recessive allels. In some natural populations different wild type allels affecting the same character were found and these wild type alleles had similarallelic dominance or they may differ in their degree of expression, that could be detected in special combinations. Such alleles are called ' Iso allels". eg. Drosphila - different dominant alleles on red eye 3 wild type alleles. They are alike in homozygous conditions and their difference appeared only in special combination.

PENETRANCE AND EXPRESSSIVITY

PENETRANCE

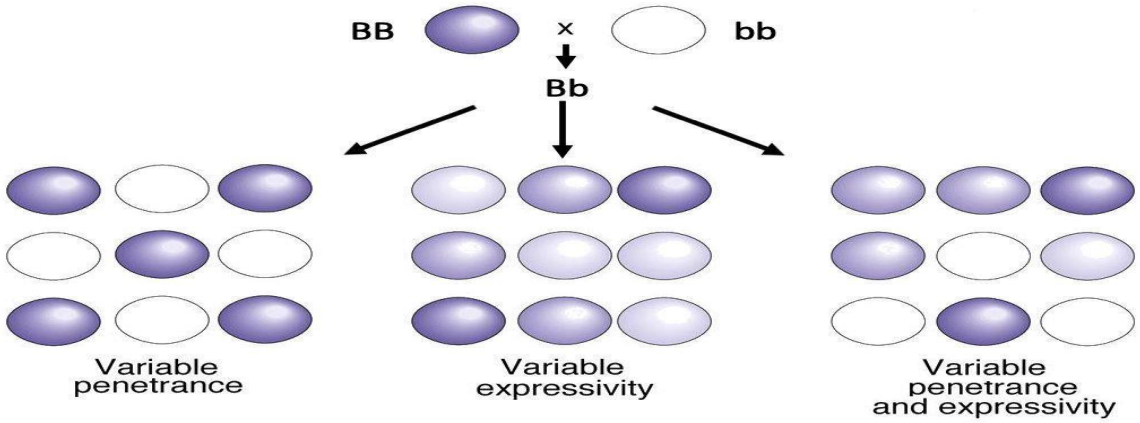
The ability of a gene to express itself in all the individuals which they are present in the appropriate genotype, this is known as complete penetrance but many genes may not produce the concern phenotype in all the individual. Such situation is known as incomplete penetrance. Eg. If a dominant gene is expressed in only 70% of the individuals, the penetrance of the gene would be 70%. Many individuals fail to do so (Incomplete penetrance

EXPRESSIVITY

The ability of a gene to produce identical phenotype in all the individual carrying in the appropriate genotype is known as uniform expressivity. The many genes produce variable phenotypes in the individual that have this gene in the appropriate genotype is called variable expressivity. The degree of phenotypic expression of a gene in the different individuals it may be uniform or variable.

For polydactyly, an extra digit may occur on one or more appendages, and the digit can be full size or just a stub. Therefore, when the P allele is present it expresses variable expressivity

1. Genotypes vary in penetrance (percent of individuals affected) and expressivity (severity of symptoms).



Expressivity of a gene is influenced by termpvating nutrition etc. The character that develop thus depend upon the genotype as well as upon the environment. It is evident that, the expression of genes depend upon the environment in which the organism develops.

PHYSICAL BASIS OF HERIDITY

Mendel had no knowledge of chromosome or genes and he was able to postulate that the inheritance in particualte and that the elements of factors controls the particular character and is transmitted from one generation to the next. His conclusions also gave the fact that;

i. Each of the two parents has two elements for a character and

ii. Only one these transmitted to the next generation through gametes.

In 1900, Sutton studied the chromosome behaviour during Meiosis and found the likeness between segregation of Mendel's factor determines during gametogenesis. It was thre fore concluded that chromosomes are the carriers of hereditary particles and the Mendel's factors are physically located in the chromosomes. In other words, he suggested that, the chromosomes constitute the physical basis of heredity. Johnnson applied the term 'gene' to represent there hereditory factors. There are handed down from parent to progeny thorugh succesive generation.

CHAPTER 6

INHERITANCE OF QUANTITATIVE TRAITS

Quantitative traits: The characters which are governed by several genes, also called polygenic traits

Qualitative traits: The characters which are governed by one or few genes, also called Oligogenic traits

Differences between polygenic and oligogenic traits

	Polygenic traits	Oligogenic traits
1.	Governed by several genes	Governed by few genes
2.	Effects of each gene is not detectable	Effect of each gene is detectable
3.	Usually governed by additive gees	Governed by non-additive genes
4.	Variation is continuous	Variation is discontinuous
5.	Separation into different classes is not possible	Separation into different classes is possible
6.	Highly influenced by environmental	Little influence by environmental factors
7.	Statistical analysis is based on mean, variances and covariance	Statistical analysis is based on frequencies or ratios

MULTIPLE FACTOR HYPOTHESIS

Polygenic inheritance quantitative characters

It shows more or less continous variation and are governed by a large number of genes called ' multiple gene' or 'multiple factor' or 'polymeric genes' or 'polygenes'.

Nilson -Ehle's studies on kernel colour in wheat

The Swedish geneticist Nilson - Ehle (1908) effected crosses between different true breeding strains of wheat with red kernels and those with white kernels. Careful examinations however revealed that, a red colour of the F1 was not so intense as the red colour of the parent and that in the F2. Some red grains wree as dark as those of parent and others only as dark as those of the F1. It was possible to separate the F2 in to the following;

Dark red	1	-	R1 R2 R2 R2 - 4 contributing genes.
Meidum dark red	4	-	3 contributing genes
Medium red	6	-	"
Light red	4	-	"
White	1	-	No "

	Red			White
Parents	R1 R1	R2 R2	x	r1 r1 r2 r2
F1		R1 r1 R2 r2 Medium red.		
F2		1 : 4 : 6 : 4 : 1		

It is evident that, red colour is due to two pairs of alleles. Each gene is capable of producing red colour. Each is in completely dominant over white and in cumulative in its effect. The intensity of red colour depends upon the number of colour producing gene present.

From these studies, Nilson-Ehle proposed the multiple factor hypothesis for the inheritance of quantitative characters. This assumes that there is a series of independent genes for a given quantitative trait. Dominance is usually in complete, but these genes are cumulative or additive in their effect. Each gene adds something to the strength of expression of the character, whereas its allele does not posses any effect.

Studies on earlength in corn (Emersona and East 1913).

Long eared sweet corn x Short eared pop corn

(mean ear length) 16.80 cm (6.63 cm)

↓

Intermediate - F1 12.12 cm

Uniform - Small variation by environment

↓

Extreme - F2 plants with longer ears than long parent.

Shorter ears that short parent.

The number of extreme types was small, large number of F2 being intermediate in ear length.

Mean length - 12.89 cm

(approximate intermediate between parents) equal to F1 means.

The increase in variability in the F2 was due to genetic segregation and recombination.

CHAPTER 7

LINKAGE AND CROSSING OVER

INTRODUCTION

A few years after the rediscovery of Mendel‟s laws of inheritance, Bateson and punnett (1905) observed in sweet pea that two pairs of alleles do not assort independently. Morgan (1910) found the same phenomenon in **Drosophila** and Hutchinson observed a clearcut case of linkage in Maize. All these researchers in found that genes inherit in group rather than individually. This tendency of two or more genes to remain together in the same chromosome during inheritance is referred to as linkage is the tendency of genes to be inherited in groups. The main features of linkage are given below:

1. Linkage involves two or more genes which are located in the same chromosome in a linear fashion.
2. Linkage may involve either dominate genes or recessive genes or some dominant and some recessive genes.
3. Linkage usually involves those genes which are located closely.
4. Presence of Linkage leads to higher frequency of parental types than recombinants in a test cross progeny. When two genes are linked the segregation ratio of a test of a cross progeny deviates significantly from the 1:1:1: 1 ratio.
5. Linkage may involve either two or more desirable traits as all undesirable traits or some desirable or some undesirable traits.
6. Linkage is observed for both oligogenic traits as well as polygenic traits. However, it is more common for the former than latter.
7. Besides pleiotropy, linkage is an important cause of genetic correlation between various plant characters.
8. The strength of linkage depends on the distance between the linked genes. Lesser the distance higher the strength and vice versa.
9. If crossing over does not occur, all the genes located in one chromosome to be inherited together. Thus the maximum number of linkage groups in an organism is equal to its haploid chromosome number.
10. Linkage can be broken by repeated intimating of randomly selected plants in segregation populations for several generations.

Linkage in maize

'C' for coloured aleurone is dominant over 'C' colourless

Sh for Full endosperm is dominant over 'sh' shrunken.

Parents (Short 'S' , 's' Col full x Colourless, Shrunken

CCSS ↓ ccss

Ce Ss Colour full

↓

F2	Colour full	7300
	Colourfull shrunken	200
	Colour full	200
	Colouless shrunken	2300

F2 did not show 9: 3: 3 : 1 ratio. There were greater number of colour full, colour shrunken (parental types) than colourfull shrunkern , colour full, If two character considered separately,t hey segregate 3 : 1

i.e .	Colour	7500	Full	- 7500
	Colouless	2500	Shrunken	- 2500

The large deviation of the observed F2 population form the excepted segregation is therefore not because the members of each pair of alleles do not segregate from each other but because of the separation in one pair of alleles is not independent of the separation in the other pair of alleles.

Test cross

Colour full x Colourless shrunken

CCSS ↓ eess

F1 CeSs x eess

↓

F2	Colour full	4800	No expected
	Col. Shrunken	200	ratio 1:1:1:1
	Col. Less full	200	
	Col less shrunken	4800	

The data show that, the two pairs of genes have nto assorted independently.

PHASES OF LINKAGE

There are two phases of linkage, *viz.*, coupling phase and repulsion phase. These phases were given by Bateson and Punett (1905), but they could not give proper interpretation of these terms. Later on, Morgan (1910) based on his studies which **Drosophila** explained that coupling and repulsion are the two aspects of the same phenomenon what we call linkage. The coupling and repulsion phases of linkage are briefly described below.

Coupling phase

The linkage between two or more either dominant (AB) or recessive (ab) alleles is referred to as coupling. A good example of coupling was reported by Hutchinson in maize for the genes governing colour of seed (coloured and colourless) and shape of seed (full and shrunken). The coloured seed is governed by dominant gene (C) and full seed is also governed by dominant gene (S). He made cross between plants having coloured full seeds (CCSS) and colourless shrunken seeds (ccss). The F1 seeds were colourless full. When the F1 was test crossed with double recessive parent the following results were obtained of 1:1:1: 1 ratio.

Repulsion Phase

The linkage of dominant allele with that of the recessive allele (AB or aB) is known as repulsion. Hutchinson also observed repulsion phases of linkage in maze. He observed this types of linkage when he made cross between plants having coloured shrunken seeds (Cs) with those having colourless fuil seed (cS). In F1 ,the seeds were coloured full. By crossing of F1 with double recessive parent the following results were obtained instead of 1:1: 1: 1: ratio.

TYPES OF LIKAGE

Linkage is generally classified on the basis of three criteria, viz., (1) Presence or absence of crossing over, (2) genes involved, and (3) the chromosome involved. These are briefly described below:

1. Based on crossing over

(1) Complete Linkage. Linkage in which crossing over does not occur is Known as complete linkage or absolute linkage. In other words when only parental types are obtained from the cross progeny, it refers to complete linkage. Good example of complete linkage is **Drosophila** male and female silk moth.

(2) Incomplete Linkage. If some frequency of crossing over also occurs between linked genes, it is known as incomplete linked. To put in other way, when only recombination is also observed in the test cross progeny, besides combinations, it refers to incomplete linkage. Incomplete linkage has been observed in maize, pea, **Drosophila** female and several other organisms.

2. Based on Genes Involved

(1) Coupling Linkage. It refers to linkage either between dominate gene or between recessive genes. Such linkage has been reported in pea, maize and several other crops.

(2) Repulsion Linkage. It refers to linkage of some dominate genes with some recessive genes. This types of linkage has also been observed in pea, maize and several other crops.

3. Based on Chromosome Involved

(1) Autosomal Linkage. It refers to linkage of such genes which are located in other than sex chromosome (autosomes).

(2) X-Chromosome Linkage. It refers to the linkage of genes which are located in sex chromosome.

LINKAGE GROUPS

Linkage groups of genes of which are present in one chromosome. In other words, all those genes which are located in one chromosome constitute one linkage group. The number of linkage groups is limited in each individual. The maximum number of linkage groups in

equal to the haploid chromosome number of an organism. However, in case of species having dissimilar sex chromosome number. For example, there are ten linkage groups in corn, 7 in garden pea, 7 in barley, 4 in **drosophila melanogaster** and 23 in man.

Genes are assigned to various chromosome of a species with the help of deletion, monatomic and nullisomic analyses. Linkage groups are assigned to different chromosome in a linear fashion and same sequence as they normally list. Generally, a relative length of various linkage groups in species exhibits a close agreement with the relative length of which they exist.

DETECTION OF LINKAGE

Test cross is the most common method of detecting the linkage. In this method, the F1 heterozygous at two loci (Say AaBb) is crossed to a double recessive parent (aabb) and the phenotypic ratio of test cross progeny is Examined. If the phenotypic ratio of test crosses progeny shown 1:1:1:1: ratio of parental and recombinant genotypes, it indicates absence of linkage. If the frequency of parental types and recombinant types deviate significantly from the normal test cross ratio of 1:1:1:1, it reveals presence of linkage between two genes under study.

There is another way to detect the presence or absence of linkage. The individual hetercrozygous at two loci (AaBb) is self-pollinated. If there is complete dominance at each locus and no epistasis, the segregation ratio of the progeny will be 9: 3:3:1. Presence of linkage either in coupling or repulsion phase will lead to significant deviation from 9: 3:3:1 ratio. The deviation of observed values from of the expected ratio is tested with the help of X2 test.

Segression of two pairs of genes on two pairs of chromosomes

Let us suppose that, gene 'C' is located on chromosome number 9 and 'S' on chromosome number 10 of maize. The segregation of chromosome bearing C and c is entirely independent of segregation of chromosome bearing S and s. So four type of gametes Cs, Cs, eS, eS are formed in F1 and F2 normal dihybrid ratio 9:3:3:1 and test cross 1:1:1:1

Segregation fo two pairs of genes on one pair of chromosomes

Let us suppose that, two genes C and S are located on chromosome No. 9 during meiosis only 2 gametes will be formed Cs and cs gametes.

So, Genes C and S situated on same chromosomes are said to be linked. Linkage is the association of character in inheritance due to fact that genes determining them are physically located on the same chromosomes.

Detectionof Linkage

Compare the number of individuals observed in each class with those expected on the basis of independent assortment and then to test the deviation between these two values by chi-square test.

Linkage Group

The number of linkage groups will be equal to the haploid number of chromosomes which the species possess. Thus maize has 10 pairs chromosomes has 10 linkage groups.

Symbol of linked genes

While representing linked gene, the two homologous chromosomes are indicated by two horizontal links.

e.g. $\frac{CS}{cs}$ $\underline{CS}$ CS/cs
cs

Coupling

In the condition is linked inheritance in which an individual heterozygous for two pairs of genes receives the two dominant member from one parent and the two recessive members from the other parent.

eg. CS / CS x cs / cs

↓

It is also said to be in C is position.

CS / cs

Repulsion CS / Cs x cS / cS

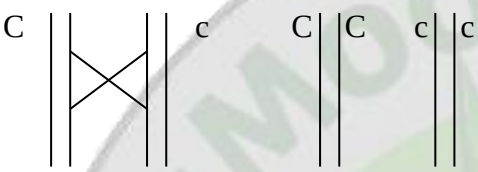
↓

Cs / cS 'Trans' position

Repulsion is the condition is linked inheritance, in which an individual heterozygous for two pairs of linked genes receives the dominant member of one pair and the recessive member of the other pair from one parent and the reverse from the other parent.

Crossing over

Leading to recombination of linked genes is due to the exchange of corresponding segments between the chromatids of homologous chromosomes and was first observed by Belgian cytologist Janssens in 1909.



Parental type CS, cs

Recombination types Cs, cS

Linkage studies revealed the following

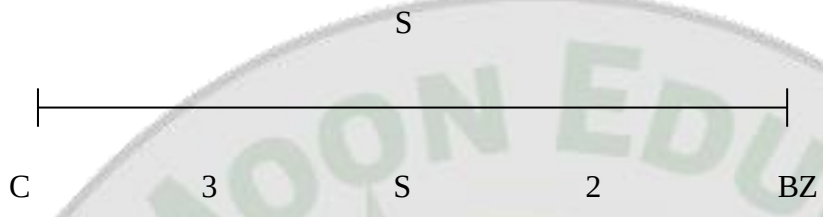
1. Genes that assort at random are non linked genes. Genes that do not segregate at random are linked genes.
2. Linked genes are arranged in a lines fashion on the chromosome. Each linked gene has a definite and constant order in its arrangement.
3. The distance between the linked genes determines the degree of strength of linkage. Closely located genes show stronger linkage that the widely located genes.
4. Linked genes do not always stay together, but are often exchanged reciprocally by cross over.

LINKAGE MAP (Cross over map / chromosome map or genetic map)

Magan postulated that genes are arranged in linear order along with length of chromosome, each gene having a fixed place on the chromosome and its allele, a correspondign position on the homologous chromosome. Under standardized environmental conditions, thre frequency of crossing over of a pair linked genes has been found to be cosntant and Magan put forward the hypothesis that it depends upon the distance between two genes on the chromosome. The greater the distance between the two genes, the greater in the chance that a Chiasma will occur between their loci, and the higher in the percentage of crossing over between them. If therefore , the percentage of crossing over between various genes are determined experimentally, the gene can be mapped in their order on the chromosome.

In mapping genes, a unit of distance must be used and this unit is called a map unit, which is the space within which one percent of crossing over takes palce. If percent of cross over between two linked genes is 1% it means that the map distance between these two linked genes is one unit of map distance or one map unit or one centimaorgan.

If the genes are in the order C, S, BZ,



The genes C and BZ show 5% crossing over . (If the gene are in the order C, BZ and Z, the genes C and BZ should show 1% corssing over. Experimental data revealed that the percentage of crossing over between C and BZ in 5. There three genes C, S and BZ on the ninth chromosome of maize and plotted as above.

Importance of linkage in breeding

When there is a close linkage between desirable and undesirable characters these genes are inherited in blocks and not individually and recombination is practically nil. In such cases linkage has to be broken by ' irradiation'.

LINKAGE AND PLEIOTROPY

A close association between two or more characters may result either due to linkage or pleiotropy or both. Pleiotropy refers to the control of two or more characters by a single gene. A tight linkage between two loci can be confused with pleiotropy. The only way to distinguish between linkage and pleiotropy is to find out a crossover product between linked characters. Intermating in segregating population may break a tight linkage, but a hung population has to be raised to find out the crossover product. If a crossover product is not found inspite of repeated intermating, there seems to be the case of pleiotropy rather than linkage.

Crossing Over and its Significance

INTRODUCATION:

Crossing over refers to the interchange of parts between non-sister chromatids of homologus chromosomes during meiotic prophase (pachytene). In other words, crossing over result from exchange of genetic material between non-sister chromatids involving breakage and precise point. The term crossing over was first used by Morgan and cattell in 1912. The main features of crossing over are given bellow:

1. Crossing over takes place during meiotic prophase, *i.e.*, during pachytene. Each pair of chromosome has four chromatids at that time.
2. Crossing over occurs between non-sister chromatids. Thus one chromatid from each of the two homologus chromosome is involved in crossing over.
3. It is universally accepted that crossing takes place at four strand stage.
4. Each crossing over involves only two of the four chromatids of two homologus chromosome. However, double or multiple crossing over may involve all four, there or two of the four chomatids, which is very rare.
5. Crossing over leads to new combinations between linked genes. Crossing over generally yields two recombinant types or crossover types and two parental types or non – crossover types.
6. Crossing over generally leads to equal segments or genes and recombination is always reciprocal. However, unequal crossing over has also been reported.
7. The value of crossover or recombinants may vary form 0-50%.
8. The frequency of recombinants can be worked out from the test cross progeny. It is expressed as the percentage ratio of recombinants to the total population (recombinants + parental types) Thus,

$$\text{Crossing over frequency (\%)} = \frac{\text{No. of recombinants}}{\text{Total progeny}} \times 100$$

In cases of two strand crossing over, somatic crossing over, sister stand crossing over and unequal crossing over"s are known. However, frequency of such cases is extremely low, *i.e.* infractions. Crossing over differs from linkage in several aspects (Table 15.1).

Table: Difference between crossing over and linkage

Crossing Over	Linkage
It leads to separation of linked genes.	It keeps the genes together.
It involve non-sister chromosome of homologous Chromosomes.	It involves individual chromosome.
Frequency of crossing over can never exceed 50%.	Linkage groups can never be more than haploid chromosome number.
It increases variability by forming new gene combinations.	It reduces variability.
It provides equal frequency of parental and recombinant types in test cross progeny.	Provides higher frequency of parental types than recombinant types in test cross progeny.

Over takes place. Chiasma was first discovered by Janssen‟s in 1909. Depending on the position, chiasma is of two types, viz., terminal and interstitial. When the chiasma is located at the end of the pairing chromatids, it is Know as terminal chiasma and when it is the middle part of non-sister chromatids, it is referred to as interstitial chiasma. Later on interstitial chiasma is changed to terminal position by the process of chiasma terminalisation. The number of chiasma per bivalent may vary from one to more than one depending upon the length of chromatids. When two chiasmate are formed, they may involve two, three or all the four chromatids.

CHAPTER 8

CYTOPLASMIC INHERITANCE

Introduction:

The inheritance of most the characters of an individual is governed by nuclear genes. But in some cases the inheritance is governed by cytoplasmic factors or genes. When the transmission of characters from parents to offspring is governed by cytoplasmic genes, it is known as cytoplasmic inheritance or extranuclear inheritance or extrachromosomal inheritance or non-mendelian inheritance or organellar inheritance. The first case of cytoplasmic inheritance was reported by Correns in 1909 in four o‟ clock (**Mirabilis Jalapa**) for leaf colour. Later on, cytoplasmic inheritance was reported by various workers in various organisms.

Important Features

Cytoplasmic inheritance differs from Mendelian inheritance in several aspects (Table 18.1) and exhibits some characteristic features. The important characteristic features of cytoplasmic inheritances are briefly described below:

1. Reciprocal Differences

Characters which are governed by cytoplasmic inheritance invariably exhibit marked differences in reciprocal crosses in F1, whereas in case of nuclear inheritance such differences are not observed except in case of sex linked genes.

2. Maternal Effects

In case of cytoplasmic inheritance, distinct maternal effects are observed. This is mainly due to more contribution of cytoplasm to the zygote by female parent than male parent. Generally ovum contributes more cytoplasm to the zygote the sperm.

3. Mappability

Nuclear genes can be easily mapped on chromosome, but it is very difficult to map cytoplasmic genes or prepare linkage map for such genes. Now chloroplast genes in **Chlamydomonas** and maize, and mitochondrial genes in human and been mapped.

4. Non-mendelian segregation

The Mendelian inheritance exhibits typical segregation pattern. Such typical segregation is not observed in case of cytoplasmic inheritance. The segregation when occurs, is different from Mendelian segregation.

5. Some Segregation

Characters which are governed by cytoplasmic genes usually exhibit segregation in somatic tissues such as leaf variegation is very rare for nuclear genes.

6. Infection –like Transmission

Cytoplasmic traits in some organisms exhibit infections like transmission. They are associated with parasites, symbionts or viruses present in the cytoplasm. Such cases do not come under true cytoplasmic inheritance.

7. Governed by Plasma genes

The true cases of cytoplasmic inheritance are governed by chloroplast or mitochondrial DNA. In other words, plasma genes are made of cp-DNA or mt-DNA.

Table: Difference between Mendelian inheritance and cytoplasmic inheritance

Mendelian Inheritance	Cytoplasmic Inheritance
1. Governed by nuclear genes.	Governed by plasma genes.
2. Exhibits distinct segregation pattern.	Does not exhibit distinct segregation.
3. Reciprocal differences are not observed.	Reciprocal differences are observed.
4. Does not show maternal effects.	Exhibits maternal effects. .
5. Genes can be easily mapped on chromosomes.	Mapping of plasma genes is very difficult.
6. Nuclear genes are associated with chromosomes.	Plasma genes are associated with either chloroplast DNA or mitochondrial DNA.

Maternal Effects

When the three expression of a character is influenced by the genotype on female parent, it referred to as maternal effect. Such characters exhibit clearcut differences in f1 for

reciprocal crosses. Maternal effects are known both in plants and animals. Some examples of maternal effects are briefly presented below.

Coiling Pattern of Shell in Snail

The effect of maternal genotype on the coiling behaviour in water snail was studied by studied by Sturtevant. There are two types of coiling pattern of shell in snail **(*Limnaea perrgra***), viz., right handed (dextral) and left handed (sinistral) . The coiling behaviour is controlled by a single gene. The dextral coiling behaviour is governed by dominant allele D and sinistral by recessive allele d. When a cross is made between dextral female and sinistral male, it produces dextral snails in F1 as well as in F2. However, F3 a segregation ratio of 3 dextral and I sinistral is observed. Similarly, when a reciprocal cross is made, i.e. sinistral as female and dextral as male, all the snails are sinistral in F1 and dextral in F2. Again in F3 a ratio of 3 dextral and I sinistral is observed (fig. 18.1). This indicates that the inheritance of coiling direction in water snail depends on the genotype of female parent and not on its own genotype.

The maternal genotype affects the organization of egg cytoplasm. In other words, it affects the orientation of first cleavage plain in the zygote. If it is tilted to the left, successive cleavages will produce a spiral to the left. If it is tilted to the right a dextral pattern will follow (Suzuki and Griffiths 1976).

1. Kappa Particles in Paramecium.

There are two types of in **Paramecium.** One has kappa particles in it cytoplasm and other does not have such particles. The presence of kappa particles in the cytoplasm leads to production of a toxin Knows paramecin. This toxin can kill the strain of **Paramecium which** lacks kappa particle. Thus, the strain with kappa particle is known as killer strain and that without kappa particle is called as sensitive strain.

Multiplication of kappa particles in the cytoplasm takes place by fission. However, their multiplication is governed by a dominant nuclear gene (K). They can multiply in the homozygous dominant (KK) or heterozygous (Kk) **individuals**. Kappa particle cannot multiply in recessive (kk) individuals. Even if kappa particle are the introduced into kk strains, they will gradually disappear dua to their inability to multiply and strain will become sensitive. Though the multiplication of kappa particles is dependent on nuclear genes, their action is independent of nuclear gene. The inheritance of kappa particles can be studied by conjugation between killer and sensitive strains. The conjugation may be of two types, via,. 1. Short duration conjugation and 2.long duration conjugation. The consequences of such

(1) **Short Duration Conjugation**. Short duration conjugation leads to exchange of nuclear genes between the killer and sensitive strains. Exchange of cytoplasm does not take place in such conjugation. Thus, the ex-conjugant‟s (resultant strains) will be heterozygous (kk) for killer gene. However, the strain with killer cytoplasm produces killer (KK) and sensitive strains by further division, whereas the sensitive stain produces only sensitive strains (kk) by further division. This clearly indicates that killer character is not governed by nuclear gene.

(2) **Long Duration Conjugation**. Such conjugation between killer and sensitive strains leads to exchange of both nuclear genes as well as cytoplasm. Here both the ex-conjugant‟s are heterozygous (kk) but killer. Auto gamy of both the ex-conjugant‟s produces killer and sensitins in 1:1 ratio. This has demonstrated that kappa particles have cytoplasmic inheritance.

Plastid Inheritance:

Chloroplasts are the important plastids. Plastids have green pigment called chloroplasts. Plastids self duplicate have some amount of DNA and play an important role in cytoplasmic inheritance. Some examples of plastid inheritance are given below:

Mirabilis Jalapa

The first conclusive evidence of cytoplasmic inheritance was reported by Correns in 1990 for left colour in four o‟ clock plant **(Mirabilis Jalapa) .**This plant has three types of leaves, viz., green, white and variegated. Three types of results were obtained from crosses between these genotypes as given below.

1. When green was used as female and eigher green, white or variegated as male, all individuals in F1 were green.
2. When white was used as female and either green, white or variegated as male, all individuals in F1 were white.
3. When variegated was used as female and either green, which or variegated as male, various proportions of green, white and variegated were obtained in F1 (Table 18.3).

   The inheritance is governed by chloroplasts which are originated from proplastids. If the proplastids are normal, they will develop into normal, chloroplasts and proplastids are mutants,.

Table: Inheritance of left colour in mirabilis Jalapa

Crosses between three leaf colours			Expression of left colour
Female		**Male**	**In F1**
Green	X	Green	Green
	X	White	Green
	X	Variegated	Green
White	X	Green	White
	X	White	White
	X	Variegated	White
Variegated	X	Green	Green, White and
	X	White	Variegated in various
	X	Variegated	**Ratios** in each cross

They will produce white chloroplasts. This suggests that green left branches have normal chloroplasts, white branches have mutant chloroplasts and variegated have a mixture of both normal and mutant chloroplasts. Since cytoplasm is contributed to the zygote mainly by female parent, the plastids are transmitted to the zygote from the female parent. These plastids are responsible for variation in the crosses of green, white and variegated leaves.

Cytoplasmic male sterility in Maize

In case of male sterility in maize, pollen grains of such male sterile are aborted. This male sterility is transmitted only through the female and never by the pollen. When all of the chromosomes of the male sterile line were replaced with chromosomes of normal plants, the line still remained male sterile, showing thereby that male sterility in controlled by some agency in the cytoplasm. It was later recognized that cytoplasmic male sterility in maize results from alterations in the heredity units in the mitochondria (mitochondrial DNA).

Inheritance of Kappa particles in Paramecium

In *Paramecium aurelia,* two strains of individuals have been reported. One is called as „ Killer‟ which secretes a toxic substance „ paramecin‟ and the other strain in known as „ sensitive‟ and is killed if comes in contact with the „paramecin‟. In the cytoplasm of the killer strain the kappa particles (cytoplasmic – DNA) are present kappa particles are absent in

sensitive strains. The transmission of kappa particles is through cytoplasm but maintenance of kappa particles and production of paramecin is controlled by „k" we assume that the killer strains carry dominant allele „kk; and that sensitive „kk".

On conjugation, conjugants exchange their nuclear material so that ex-conjugants „kk" resulted from conjugants „kk" and „kk" when conjugation is for normal time, then only nuclear material is exchanged and therefore killer will produce killer daughters and sensitive will produce sensitive daughters. But if the conjugation is in longer period, there will be exchange of cytoplasm resulting in the inheritance of kappa particles by both the ex-conjugants so that all the daughter paramecia produced are killers because all in herit the kappa particles through the mixing of cytoplasm. Therefore this trait is transmitted through cytoplasmic heredity. The trait is only stable is killer strains.

Inheritance through mitochondria

Mitochondria can self-replicate and represent another genetic system in the cell. Of course, the amount of mitochondrial DNA is so small, representing less than 1% of the nuclear DNA is mammalian cells and it can code for a part of the protein in the mitochondria. The synthesis of the cytochrome found inmitochondria for example, is known to be present in minute amount in cytoplasm under the control of nuclear genes. Therefore, it is suggested that both mitochondria and chloroplast seem to have a semi-autonomous existence and their DNA forms the basis for genetic systems separate from that in the nucleus.

CHAPTER 9

SEX DETERMINATIONAND SEX LINKAGE

In the previous classes, we focused on the traits and their inheritance pattern which were presents on autosomes. Gene expression and their inheritance present on allosome are different than autosomal genes. Those allosomes are called sex chromosome that determines the sex expression of an individual. The traits governed by the genes present on sex chromosome are called sex linked traits. The linkage observed between the genes on sex chromosome also known as sex linkage.

In human, two allosomes represents as X and Y chromosome are presents. Female are homogametic (XX) and male is heterogametic (XY). Whereas in birds, sex determining system is called ZZ/ZW; where male is homogametic (ZZ) and female is heterogametic (ZW). In drosophila the ratio of X and autosomes are determines the sex expression.

Sex determination in Drosophila:

The investigations on drosophila by C B Bridges (1925) showed that female determinates were located on the X chromosomes and male determinates were on the autosomes. Female determining genes were shown to be carried on the X chromosomes and male determining shown to be located on the three autosomal chromosomes of drosophila. The genetic balance theory f sex determination was devised to explain the mechanics of sex determination in drosophila. Bridges experimentally produced various combination of x chromosomes and autosomes in drosophila and deduced from comparison that one X chromosome and two sets of autosome produced a normal male. Normal males had a ration of x-chromosomes and two set of autosomes of 0.5. This combination of one X and two A"s resulted in a normal diploid male; the combination of two X-chromosomes and two sets of autosomes (2X + 2A, ration of 2:2=1) produced a normal diploid female.

Table: Ratio of X-chromosome to autosomes and expression of sex types in drosophila

X chromosome and set of autosomes	Ration of X/A	Sex expression
1X & 2A	0.5	Male
2X & 2A	1.0	Female

3X & 2A	1.5	Super female
4X & 3A	1.33	Super female
4X & 4A	1.0	Tetraploid female
3X & 3A	1.0	Triploid female
3X & 4A	0.75	Intersex

Sex liked inheritance:

The first extensive experimental evidence for sex linkage in a particular species came in 1910 with the discovery by T H Morgan of a white eyed mutant if drosophila. A gene had undergone a change that resulted in a phonotypic alteration. This change expressed itself as a white eye rather than the normal red eyes. The white eyed male first discovered was mated with a red eyed female. The F1 flies were all red eyed, but the F2 included both red and white in the proportion of about red to one white. All white eyed flies in the F2 generations however were males. About half of the F2 males were white eyed and half were red eyes. But all females were red eyes. In this experiment, the recessive allele was expressed only in males. Morgan arrived at an explanation by associating this gene with the X-chromosome.

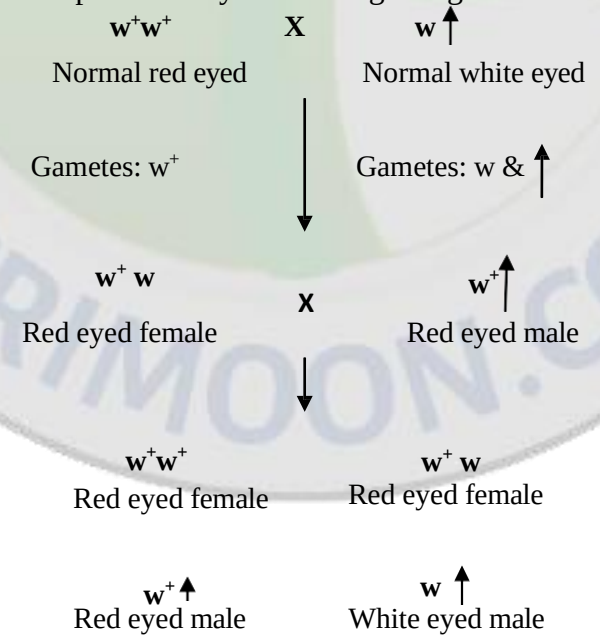


Fig: Illustration showing the Morgan'S experiment for Sex linked traits

Sex influenced dominance:

The dominance of alleles may differ in geterozygotes of the two sexes. This phenomenon is called sex influenced dominance. Gene products of heterozygotes in the two sexes may be influenced differentially by sex hormones. For example autosomal genes responsible for horns in some breed of sheep may behave differently in the presence of male and female sex hormones. Among Dorset sheep both sexes are horned and the gene for the horned condition is homozygous (HH) in all animals of the breed. In Suffolk sheep, neither sex is horned and the genotype is hh. Among the heterozygous F1 progeny from crosses between these two breeds, horned males and hornless females are produced. Because both sexes are genotypically alike (Hh), the gene must have behave as a dominant in males and as a recessive in females. That is only one allele is required for an expression in the male but the allele must be homozygous for expression in the female.

Table: Expression of h allele in sex influenced inheritance in sheep.

Genotypes	Males	Females
HH	Horned	Horned
Hh	Horned	Hornless
hh	Hornless	Hornless

Sex limited gene expression:

One sex may be uniform in expression of a particular traits and yet a transferred genes that produced a different phenotypes in offspring of the other sex. This is called sex limited gene expression. Sex hormones are apparently limiting factors in the expression of some genes. Other factors may also be involved in controlling the expression of sex limited characters. Milk production in mammals, for examples is limited to females, but certain bulls are in great demand among dairy breeders artificial inseminations and associations because their mother and daughters have increased milk production records. Another classic example is so called cock feathering in many different birds.

CHAPTER 10

MUTATION: CLASSIFICATION AND INDUCTION

Introduction:

Mutation refers to sudden heritable change in the phenotype of an individual. In molecular term mutation is defined as the permanent and relatively rare change in the number or sequence of nucleotides. Mutation was first discovered by Wright in 1791 in male lamb which had short legs. Later on mutation was reported by Hugo de Vries in 1900 in **Oenothera,** Morgan (1910) in **Drosophila** (white eye mutant 0 and several others in various organisms. The term mutation was coined by de Vries.

CHARACTERISTICS OF MUTATIONS

Mutations have several characteristic features. Some of the important characteristics of mutations are briefly presented below:

1. **Nature of change**

   Mutations are more or less permanent and heritable change in the phenotypes of an individual. Such changes occur due to alteration in number, kind or sequence of nucleotides of genetic material, *i.e.*, DNA in most of the cases.

2. **Frequency**

   Spontaneous mutations occur at a very low frequency. However, the mutation rate can be enhanced many fold by the use physical and chemical mutagens. The frequency of mutation for a gene is calculated as fellows.

   $$\text{Frequency of gene mutation} = \frac{M}{M+N}$$

   Where,

   M = number of individual expressing mutation for a gene, and

   N = number of normal individual in a population

3. **Mutation Rate**

   Mutation rate varies from gene to gene. Some genes exhibit high mutation rate than others. Such genes are known as mutable genes, *e.g.*, White eye in Drosophila. In some genes enhance the natural mutation rate of the genes. Such genes are termed as mutator genes. The example of mutator gene is dotted gene in maize. In some cases, some genes decrease the frequency of spontaneous mutations of other genes in the

same genome, which are referred to antimutator genes. Such genes have been reported in bacteria and bacteriophages.

4. **Direction of Change**

   Mutations usually occur form dominant to recessive allele or wild type to mutant allele. However, reveres mutations are also known e.g., notch wing and eye in Drosophila.

5. **Effects:** Mutations are generally harmful to the organism. In other words, most of the mutations have deleterious effects. Only about 0.1% of the induced mutations are useful in crop improvement. In majority of cases, mutant alleles have pleiotropic effects. Mutations give rise to multiple alleles of a gene.

6. **Site of Mutation**

   Muton which is a sub-division of gene is the site of mutation. An average gene contains 500 to 1000 mutational sites. Within a gene some sites are highly others. These are generally referred to as hot spots. Mutations may occurs in any tissue of an organism, *i.e.*, somatic or gametic.

7. **Type of Event**

   Mutations are random events. They may occur in any gene (nuclear or cytoplasmic), in any cell (somatic or reproductive) and at any stage of development of an individual.

8. **Recurrence**

   The same type of mutation may occur repeatedly or again and again in different individuals of the same population. Thus mutations are of recurrent nature.

CLASSIFICATION OF MUTATIONS

Mutations can be classified in various ways. A brief classification of mutations on the basis of 1.Source 2.direction 3.tissue 4. Effects 5.Sits 6.character and 7. Visibility is presented in Table 22.1.

Table: Classification and brief description of mutation

Basis of classification and types of mutation	**Brief Description**
1. Based on Source	
Spontaneous	Mutations that occur in nature
Induced	Mutation which are produced by the use of mutagenic agents.
2. Based on Direction	

Forward mutation	Any change from will type‟s allele.
Reverse mutation	A change from mutant allele to wild types.
3. Based on Tissue	
Somatic mutation	A mutation in somatic tissue.
Germinal mutation	A mutation in germ line.
4. Based on Survival	
Lethel	A mutation which kills all the individual that carries it.
Sub-lethal	When mortality is more than50%of individuals that carry mutation.
Sub-vital	When mortality is less than50%of individuals that carry mutation
Vital	When all mutant individuals survive.
5. Based on site	
Nuclear mutation	A mutation in nuclear gene.
Cytoplasmic mutation	A mutation is cytoplasmic gene.
6. Based on Character	
Morphological	A mutation that alters morphological of an individual.
Biochemical	A mutation that alters biochemical function of an individual.
7. Based on Visibility	
Macro-mutations	Mutations with distinct morphological changes in phenotype. Generally found in qualitative characters.
Micro-mutation	Mutation with invisible phenotypic changes. Generally observed in qualitative characters

KINDS OF MUTANTS

The product of mutations is known as mutant. It may be a genotype or a cell or a polypeptide. There are four main classic of identifiable mutant, *viz.*, 1.Morphology, 2.Lethal, 3.Conditional and 4.Biochemical. These are briefly described below:

Morphological

Morphological mutants refer to change in from *i.e.*, shape, size and colour. Albino spores in **Neurospora**, curly wings in **Drosophila,** dwarf peas, short legged sheep are some examples of morphological mutants.

Lethal

In this class, the new allele is recognized by its mortal or lethal effect on the organism. When the mutant allele is lethal all individuals such allele will die; but when it is semi lethal or sub-vital some of the individuals will survive.

Conditional Lethal

Some alleles produce a mutant phenotype under specific environmental conditions. Such mutants are called restrictive mutants. Under other conditions they produce normal phenotype and are called permissive. Such mutants can be grown under permissive and then be shifted to restrictive conditions for evaluation.

Biochemical Mutant

Some mutants are identified by the loss of a biochemical function of the cell. The Can assume normal function, if the medium is supplemented with appropriate nutrients. For example, adenine auxotroph can be grown only if adenine is supplied, whereas wild type does not require adenine supplement.

MUTAGENS

Mutagens refer to physical or chemical agents which greatly enhance the frequency of mutations. Various radiations and chemicals are used as mutagens. Radiations come under physical mutagens. A brief description of various physical and chemical mutagens is presented below:

PHYSICAL MUTAGENS

Physical mutagens include various types of radiations, viz., X-rays, gamma rays, alpha particles, beta particles, fast and thermal (slow) neutrons and ultra violet rays (Table 22.2) .A brief description of these mutagens is presented below:

Table: Commonly used physical mutagens (radiation), their properties and mode of action

Type Radiation	Main properties	Mode of action or changes caused
X-rays	S.I. , penetrating and non-particulate	Induce mutation by forming free radicals and ions. Case addition, transitions and transversions.
Gamma	S.I., very penetrating and non-particulate	Induce mutations by ejecting atoms from the tissue. Cause all types of changes as above.
Alpha particies	D.I., particulate, lest penetrating and positively charged.	Act by ionization and excitation. Cause chromosomal and gene mutations.
Beta Rays Particles	S.I., particulate more penetrating than alpha particles and negatively charged.	Act by ionization and excitation. Cause chromosomal and gene mutations.
Fast and Thermal Neutrons	D.I., particulate, neutral particles, highly penetrating.	Cause chromosomal breakage and gene mutations.
Ultra Violet Rays	Non-ionizing, low penetrating	Cause chromosomal breakage and gene mutations.

Note : Particulate refers to particle emitting property D I =Densely ionizing, SI=Sparsely ionizing

Table: Some commonly used chemical mutagens and their mode of action

Group of mutagen	Name of Chemical	Mode of action
Alkylating Agents	Ethyl methane Sulphonate Methyn Methane Sulphonate Ethyl Ethane Sulphonate Ethylene Imines	AT ↔ GC Transitions AT ↔ GC Transitions
Base Analogues	5 Bromo Uracil 2 Amino Purine	AT ↔ GC Transitions AT ↔ GC Transitions

Acridine Dyes	Acriflavin, Proflavin.	Deletion, addition and frame shifts.
Others	Nitrous Acid Hydroxylamine Sodium Azide	AT ⟷ GC Transitions GC→ AT Transversion AT ⟷ GC Transitions

Detection of Mutation

CIB Method in Drosophila

This method was developed by Muller for detection of induced sex Linked recessive lethal mutations in **Drosophila** male. In this technique, C represents a paracentric inversion in large part of X-chromosome which suppresses crossing over in the inverted portion. The I is a recessive lethal. Females with lethal gene can survive only in heterozygous condition. The B stands for bar eye which acts as a marker and helps in identification of flies. The I and B are inherited together because C does not allow crossing over to occur between them. The males with CIB chromosome do not survive because of lethal effect. The important steps of methods are as follows:

1. A cross is made between CIB female and mutagen treated male. In F_1, half of the males having normal X-chromosome will survive and those carrying CIB chromosome will die. Among the females, half have CIB chromosome and half normal chromosome (Fig. 23.1). From F_1 females with CIB chromosome and male with normal chromosome are selected for further crossing.
2. Now a crossing is made between CIB female and normal male. His time the CIB female has one CIB chromosome and one mutagen treated chromosome received from the male in earlier cross. This will produce two types of females, viz,. Half with CIB chromosome and half with mutagen treated chromosome (with normal phenotype). Both the progeny will survive. In case of males, half with CIB will die and other half have mutagen treated chromosome. If a lethal mutation was induced in mutagen treated X-chromosome, the remaining half males will also die, resulting in absence of male progeny in the above cross. Absence of male progeny in F_2 confirm the induction of sex linked recessive lethal mutation in the mutagen treated **Drosophila** male.

CHAPTER 11

PROOF OF DNA AND RNA AS THE GENETIC MATERIALS

Deoxyribonucleic acid (DNA) was first described by Friedrich Miescher in 1869, only four years after Mendel‟s work was published. But it took over 80 years for its role as the genetic material of most organisms to become firmly established. DNA was first characterized as acid-precipitable material from the cell nuclei of pus and fish sperm. The proportion of nitrogen and phosphorus was very unusual compared to other known organic substances, convincing Miescher he had discovered a new biological substance. He called it nuclein, because it was associated exclusively with the nucleus. Further work demonstrated that nuclein is a complex of protein and DNA.

Although clear experiments linking DNA to heredity were not performed until the mid 1940s, there was a good deal of circumstantial evidence that this was the case. In higher organisms, DNA was found almost exclusively in the chromosomes. The histone proteins and RNA, which chromosomes also contain, did not seem likely candidates as genetic material; sperm contained almost no RNA, and the histones are replaced in sperm by a different protein, protamine. Unlike RNA and protein, every diploid cell of an organism has about the same amount of DNA. In the hen, for example, the red blood cells contain 2.6 x 10-12g of DNA per cell, the kidney contains 2.3 x 10-12g per cell, and the liver contains 2.6 x 10-12g per cell. Furthermore, the amount of DNA seems correlated with chromosomal division; entering mitosis, the amount of cellular DNA doubles, while the haploid products of meiosis have only half the normal amount (thus rooster sperm contains 1.3 x 10-12g of DNA). In polyploid plants, which contain multiples of the diploid number of chromosomes, the quantity of DNA is also a multiple of the diploid amount. Thus, the close association of DNA with chromosomes strongly implicates DNA as the genetic material.

Experiments that proved DNA to be the genetic material

In 1928, Frederick Griffith was able to transform harmless bacteria into virulent pathogens with an extract that Oswald Avery proved, in 1944, to be DNA. In 1952, Martha Chase and Alfred Hershey used radioactively labeled virus DNA to infect bacteria, proving the same point. These important experiments established that DNA is the genetic material.

Experiment by Frederick Griffith

The first evidence that DNA is the hereditary material came from Frederick Griffith‟s studies in 1928. Griffith used chemical mutagens to isolate a non-virulent form of the bacterium that causes pneumonia, *Diplococcus pneumoniae*. Virulence required the presence of a polysaccharide capsule around the bacterium. The non-virulent mutants lacked this capsule. Colonies of non-virulent, capsule-less bacteria appeared rough and were designated R. In contrast, the virulent form produced colonies that appeared smooth, so it was designated S. Several virulent forms were known, each with a characteristic polysaccharide capsule (called IS, IIS, IIIS, etc.), which is genetically inherited and is immunologically distinct from other forms. A smooth bacterium of a particular capsule type (say IIS) can mutate to a non-encapsulated, non-virulent form (IIR, because it derives from a type II cell). This happens at a very low frequency (in less than one in a million cells), but it is inherited when it does occur. Similarly, the IIR cell can mutate back to the IIS virulent form at low frequency. However, the IIR cell line cannot mutate to a IIIS virulent form. This property provides the key to the experiment.

Griffith mixed Pneumococcus type IIR with IIS cells that had been killed and rendered non-virulent by heating them to 65°C, and he injected them into a host rabbit or, in other experiments, into a mouse. None of the strains injected alone produced disease, and no disease was expected from the mixed injections, as neither strain was virulent. However, many of the rabbits given mixed injections did come down with pneumonia and died. When analyzed, they all contained living virulent type IIIS cells! These cells could not have arisen from the type IIR cells by mutations (they would have produced type IIS cells), and the type IIIS cells were demonstrably dead (injected alone they caused no disease). Some factor must have passed from the dead IIIS cells to the live IIR ones, endowing them with the ability to make a capsule of the III type. Griffith called the factor “transforming principle” and the process genetic transformation (figure 6.1). The transforming principle could be isolated as a cell-free extract and was fully active. The stability of the principle‟s transforming activity to heat treatment at 65°C suggested that it was not a protein (such high temperatures denature most proteins).

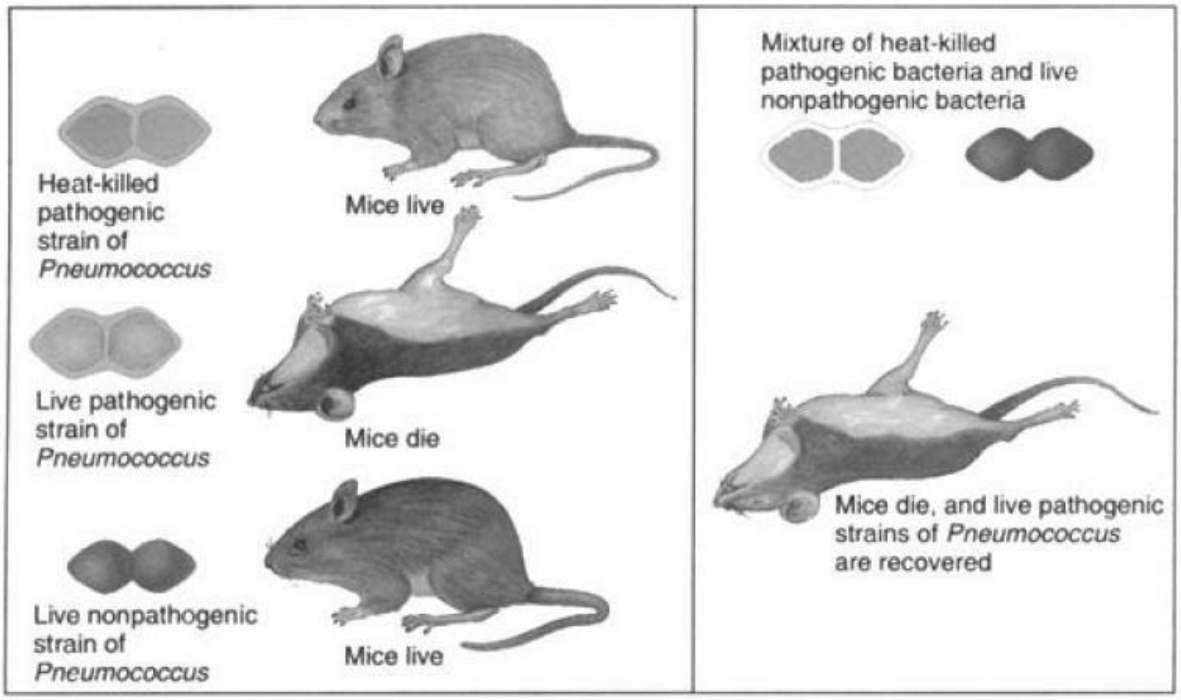


Figure: Griffith's discovery of the "transforming principle"

Experiment by Oswald Avery, C. M. MacLeod, and M. J. McCarty

In 1944, Oswald Avery, C. M. MacLeod, and M. J. McCarty succeeded in isolating a highly purified preparation of DNA from the type IIIS bacteria. The preparation of this type IIIS DNA was fully active as a transforming agent and could transform type IIR cells into type IIIS cells in a test tube. If the DNA was destroyed by deoxyribonuclease (an enzyme that specifically attacks DNA), all transforming activity was lost. It therefore seemed clear that DNA was "functionally active in determining the biochemical activities and specific characteristics of pneumococcal cells." These experiments by themselves, however, do not establish that DNA is itself the genetic material. Perhaps DNA acts upon the genetic material of the recipient cell changing its genes to resemble the genes of the DNA donor? A clear demonstration was provided by experiments on bacterial viruses.

Experiment by Alfred Hershey and Martha Chase

The experiment that clearly linked DNA and heredity were those performed by Alfred Hershey and Martha Chase in 1952 (figure 6.2). They chose to explore the genetic properties of DNA using bacterial viruses. Viruses are small, very simple aggregates of nucleic acid and protein. Several types of viruses attack bacteria and are known as bacteriophages (literally: "bacteria-eaters"). One of the viruses that attack the bacterium *Escherichia coli* is the

bacteriophage T2. It contains only protein and DNA; the DNA forms the central core of the virus, while the protein surrounds the core like a coat. Phages infect bacteria by adsorbing to the cell walls and injecting the genetic material into the bacteria. This material causes the production of many new viruses within the cell. Eventually the cell is ruptured (lysed), and the new viruses are released.

The chemical make-up of protein and of DNA is quite different. Hershey and Chase used these differences to distinguish between them. DNA contains phosphorus and proteins do not; proteins, on the other hand, usually contain sulfur, and DNA does not. By specifically labeling the phosphorus and sulfur atoms with radioisotopes, Hershey and Chase could distinguish unambiguously between the protein and the DNA of the phage and determine whether either or both were injected into the bacterial cell during the course of infection. When bacteriophage labeled with ^{32}P DNA were allowed to infect a cell, almost all the label entered the cell. If such infected cells were allowed to lyse, the label was found among the progeny viruses.

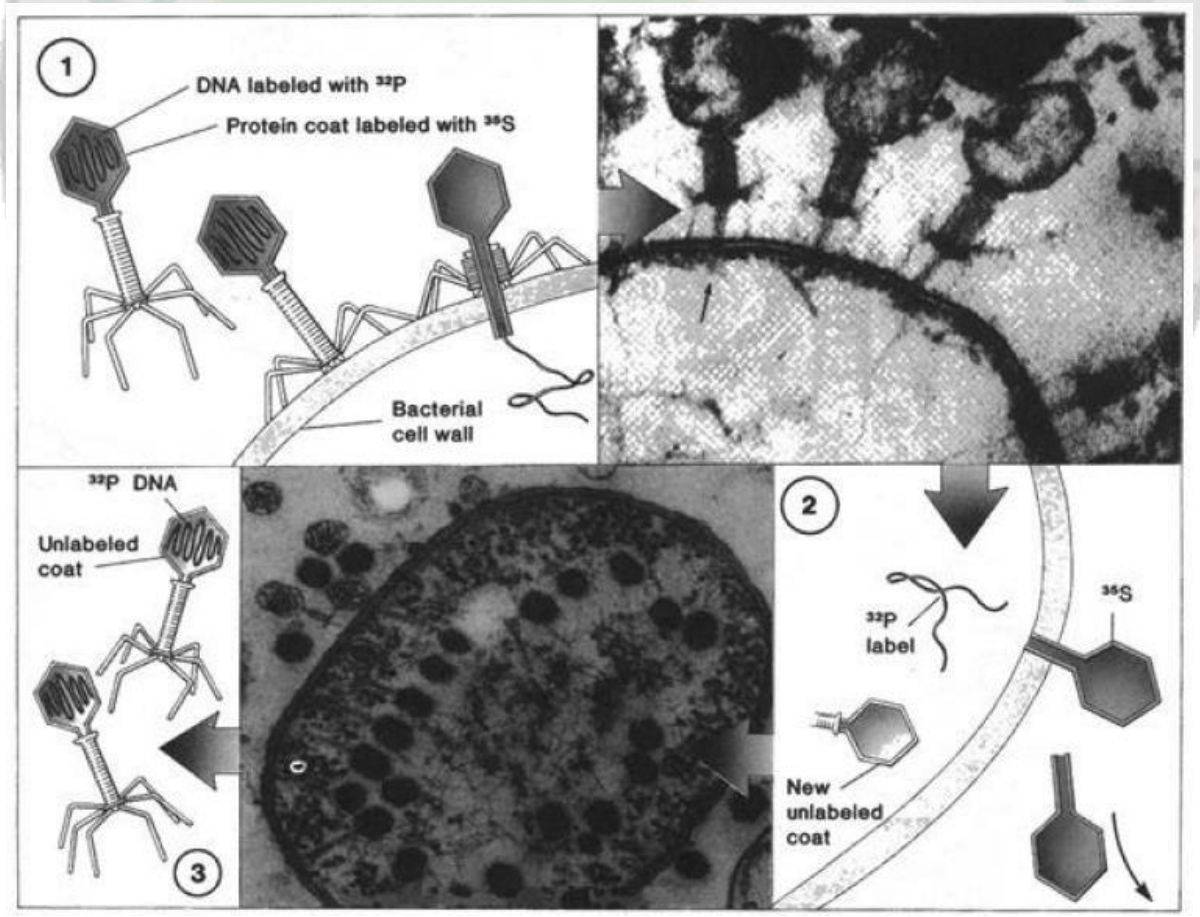


Figure: Experiment conducted by Alfred Hershey and Martha Chase

The opposite occurred when ^{35}S-labeled phage infected a bacterial culture. Almost all label remains on the outside of the bacterium, bound to fragments of the cell wall. A small amount of protein did enter the bacterial cell in the course of infection. That this was not involved in the production of new bacteriophage could be demonstrated by repeating the experiment with bacteria stripped of their cell walls (protoplasts). If protoplasts were infected with ^{32}P phage DNA free of protein, virulent phages were produced. If the purified ^{32}P was first treated with DNAase, no progeny phages were produced. Clearly the labeled DNA contained all the information necessary to produce new virus particles.

Proof of RNA as the genetic material

Some viruses do not contain DNA, being made up instead of protein and RNA (ribonucleic acid). The tobacco mosaic virus (TMV) is such an RNA virus. H. Fraenkel-Conrat and others were able to dissociate the TMV into its constituent protein and RNA parts (figure 6.3). When the parts were mixed, they reformed TMV particles that were normal in every respect. That the RNA contained the genetic information was demonstrated by isolating protein and RNA from several different types of TMV, with subsequent combinations of protein and RNA mixed together.

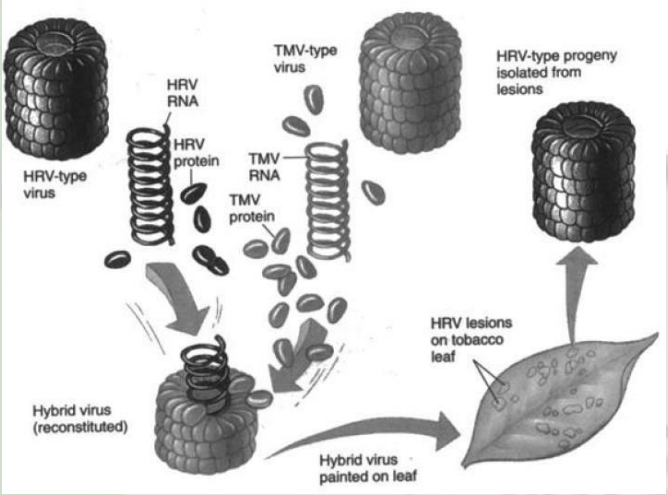


Figure: Experiment conducted to prove RNA as genetic material in TMV

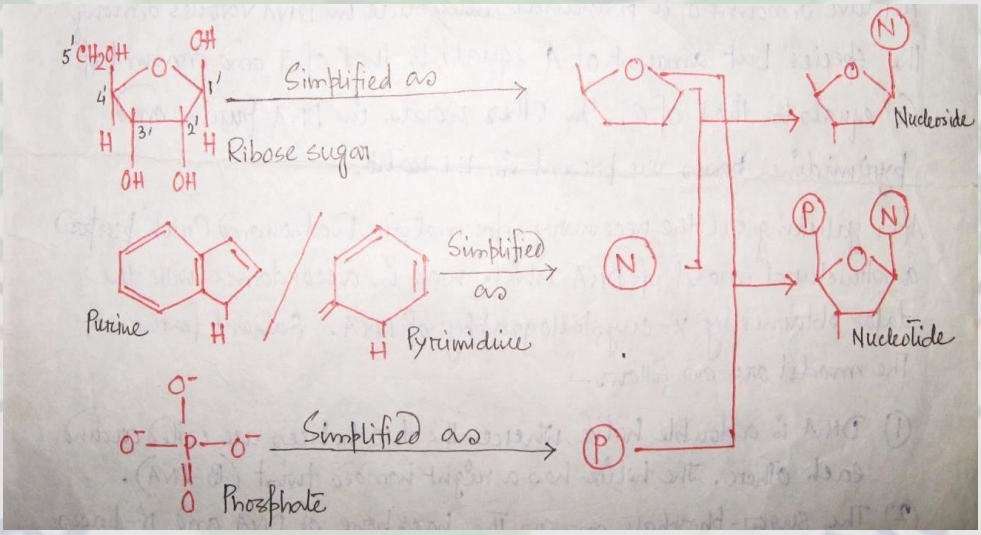
These reconstituted viruses, containing protein from one type and RNA from another, were then allowed to infect tobacco cells. In every case the progeny TMVs proved to have the protein coats of the type that had contributed the RNA, and not of the type that had contributed the protein. Thus, in the tobacco mosaic virus, the RNA, rather than the protein, must be acting as the genetic material.

CHAPTER 12

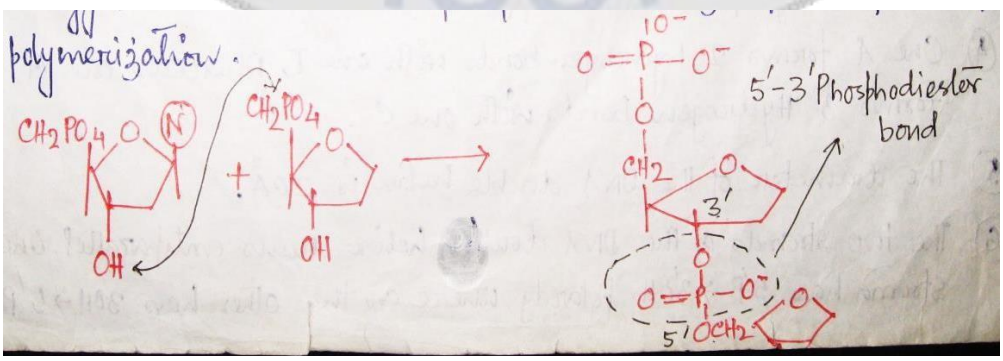
STRUCTURE AND FUNCTION OF DNA

DNA structure

With the discovery of nucleic acids (DNA and RNA) as genetic materials, much attention was given on discovering their chemical nature and structure. Nucleic acids are basically polymers of nucleotides. A nucleotide is composed of a ribose sugar (deoxy ribose sugar in case of DNA), a phosphate group and a nitrogenous base. The nitrogenous base may be purine or pyrimidine. Two types of purines (adenine and guanine) and 3 types of pyrimidine (cytosine, thymine in case of DNA and uracil only in case of RNA) are found. The ribose sugar along with the base is called a nucleoside. Hence, nucleotide can be termed as nucleoside phosphate.



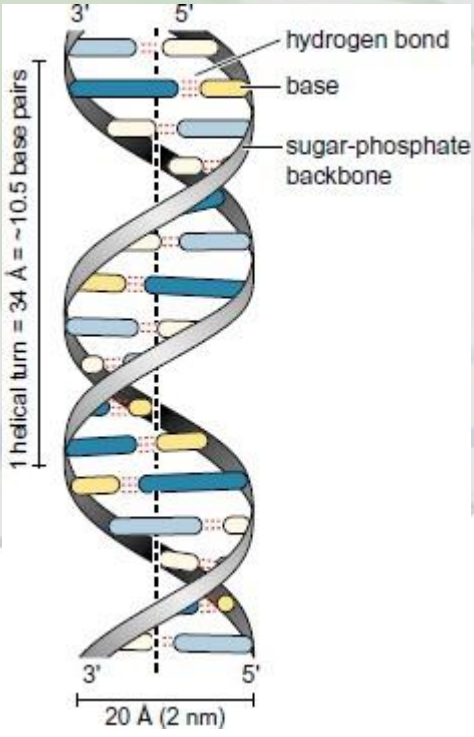
The two adjacent nucleotides are joined by 5‟-3‟ phosphodiester bond. In the cell nucleotides remain as nucleoside triphosphates. The energy held in the extra 2 phophates is utilized for the process of polymerization.



The double helical structure of DNA:

The structure of DNA mleculae was solved by James Watson and Francis Crick in the year 1953. For the same, Watson , Crick and M. Wilkins received the Nobel Prize in the year 1962. Solving the structure of DNA was not at all an easy task. In the year 1953 only, Linus Pauling was busy in proposing a 3 stranded structure of DNA. However, Watson and Crick emphasised on the X-ray crystallography of DNA, as well as the finding of Erwin Chargaff, which helped them to find out the actual structure of DNA. Before Chargaff, scientists believed on the tetra-nucleotide hypothesis which claimed that in DNA A,G,C and T are present in equal proportion. However, Chargaff found that relative amount of a particular nucleotide in DNA varied among the species. But amount of A equals to that of T and amount of G equals to that of C. In other words, in DNA, purine and pyrimidine bases are present in 1:1 ratio.

After gathering all the necessary information, Watson and Crick proposed a structural model of DNA. , which was in accordance with the data obtained by X-ray crystallography of DNA. Salient features of the model are as follows:



1. DNA is a double helix, where the two helices are coiled around each other. The helix has a right handed twist (B DNA)

2. The sugar phosphate makes the backbone of the DNA and N bases are projected inside.

3. In order to keep the diameter of the helix constant, one purine base always pairs with a pyrimidine base.

4. One A forms 2 H bonds with one T, whereas One G forms 3 H bonds with one C.

5. The diameter of the DNA double helix is 20Å.

6. The 2 strands of the DNA double helix runs anti-parallel. One strand has 5‟P to 3‟OH polarity, whereas the other strand has 3.OH to 5‟P polarity.

7. The length of one turn (pitch) in DNA double helix is 34 Å.

8. In a complete turn, approximately 10 nucleotides are present in one strand.

9. The distance between 2 adjacent nucleotides in a strand of DNA is thus ~3.4 Å.

10. The angular distance or twist between 2 adjacent nucleotides in DNA is thus ~ 36°.

11. The double helical structure of DNA contains major grooves and minor grooves, where the similarity axis passes through the major groove only.

Other forms of DNA:

Watson and Crick solved the structure of B form DNA. However, 2 other forms (A and Z) of DNA are also found. The major differences between these forms are as follows-

	Helix Type		
	A	B	Z
Overall proportions	Short and broad	Longer and thinner	Elongated and slim
Rise per base pair	2.3 Å	3.32 Å	3.8 Å
Helix-packing diameter	25.5 Å	23.7 Å	18.4 Å
Helix rotation sense	Right-handed	Right-handed	Left-handed
Base pairs per helix repeat	1	1	2
Base pairs per turn of helix	~11	~10	12
Rotation per base pair	33.6°	35.9°	−60° per 2 bp
Pitch per turn of helix	24.6 Å	33.2 Å	45.6 Å
Tilt of base normals to helix axis	+19°	−1.2°	−9°
Base-pair mean propeller twist	+18°	+16°	~0°
Helix axis location	Major groove	Through base pairs	Minor groove
Major-groove proportions	Extremely narrow but very deep	Wide and of intermediate depth	Flattened out on helix surface
Minor-groove proportions	Very broad but shallow	Narrow and of intermediate depth	Extremely narrow but very deep
Glycosyl-bond conformation	anti	anti	anti at C, syn at G

CHAPTER 13

REPLICATION OF DNA AND ITS REPAIR

DNA replication is copying genetic information for transmission to the next generation. It occurs in S phase of cell cycle. Process of DNA duplicating itself begins with the unwinding of the double helix to expose the bases in each strand of DNA. Each unpaired nucleotide will attract a complementary nucleotide from the medium which will form base pairing via hydrogen bonding. Enzymes (DNA polymerase) link the aligned nucleotides by phosphodiester bonds to form a continuous strand.

DNA replication is semiconservative in nature

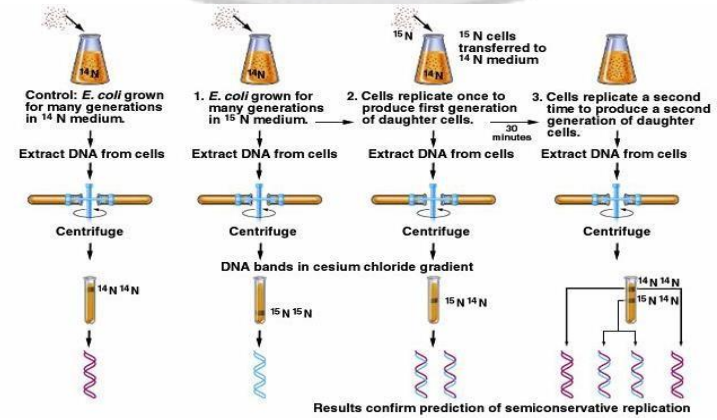
Theoretically DNA replication can occur in 3 modes:

A. Semiconservative replication: which supports the Watson and Crick model

B. Conservative replication: here the parental double helix remains intact; both strands of the daughter double helix are newly synthesized

C. Dispersive replication: here, at completion, both strands of both double helices contain both original and newly synthesized material.

The experiment conducted by Meselson and Stahl clearly proved the DNA replication to be semiconservative in nature. Bacteria were grown in media containing either normal isotope of nitrogen (14N) or the heavy isotope (15N). DNA banded after equilibrium density gradient centrifugation at a position which matched the density of the DNA: heavy DNA was at a higher density than normal DNA. When bacteria grown in 15N were transferred to normal 14N containing medium, he newly synthesized DNA strand had the 14N while the parental strand had 15N. They checked the composition of the resulting DNA molecules by density gradient centrifugation, and found an intermediate band, indicating a hybrid molecule containing both 14N and 15N DNA.

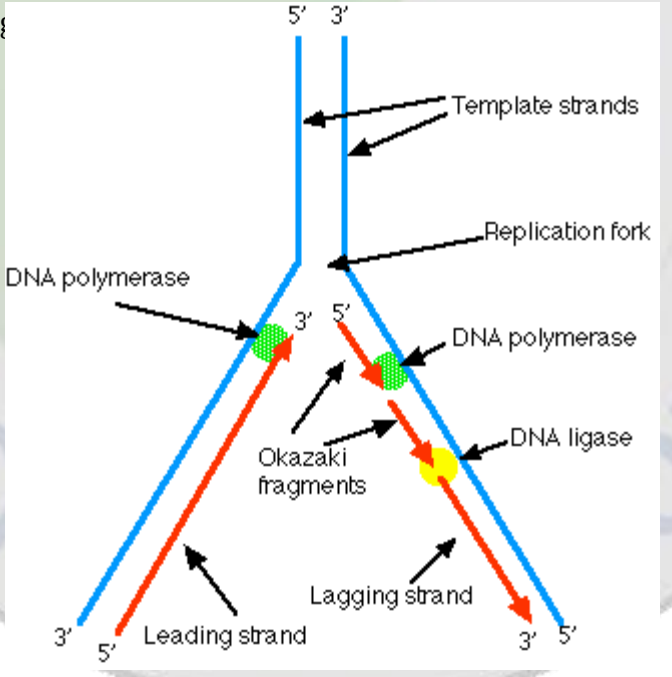


Basics of DNA replication:

DNA replication is a controlled process,that occurs at specific times during the cell cycle. It requires a set of proteins and enzymes, and requires energy in the form of ATP. There are two basic steps: 1. Initiation and 2. Elongation. At the same time, there are two basic components: 1. Template and 2. Primer.

DNA polymerase is the enzyme that extends the primer. The DNA Pol III produces new stands of complementary DNA, whereas DNA Pol I fills in gaps between newly synthesized Okazaki segments. Some additional enzymes/proteins required include i) DNA helicase that unwinds double helix, ii) Single-stranded binding proteins that keep helix open iii) Primase that creates RNA primers to initiate synthesis and iv) Ligase that welds together Okazaki fragments.

DNA replications starts in one strand (the leading strand) and proceeds continuously, whereas replication of the other strand lags behind (lagging strand) and produces discontinuous fragments (Okazaki fra



Some key points are:

1. DNA replication is semi-conservative.

2. One strand from each of the initial two strands end up in a daughter strand.

3. Each strand serves as a template for a new strand

4. New strand is formed by complementary base-pairing of the correct nucleotide plus formation of a phosphodiester bond

5. Synthesis begins at replication origins -about 100 nucleotides long rich in A-T, which are easier to pull apart because have 2 rather than 3 hydrogen bonds

6. Initiator proteins bind at replication origins and recruit DNA replication machinery proteins

7. DNA polymerase is responsible for catalyzing synthesis of new strands

8. Replication forks form and involve a leading and a lagging strand

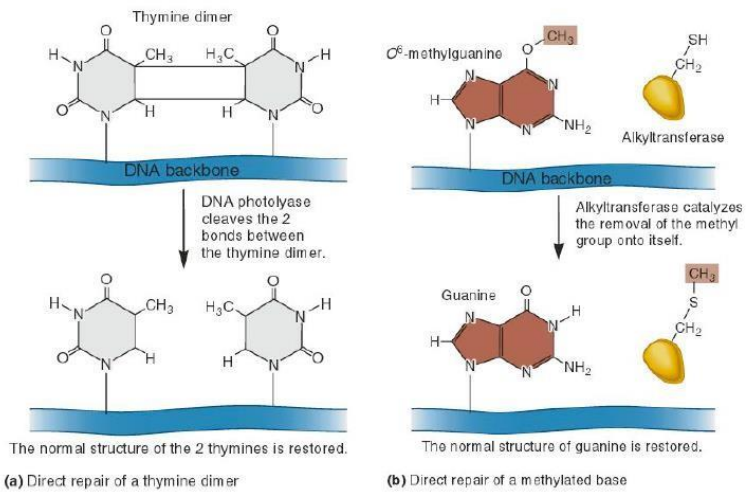
9. Replication of DNA is directional; two strands are antiparallel

10. DNA polymerase can only synthesize from 5‟ to 3‟ direction, adding new nucleotides to the 3‟ end.

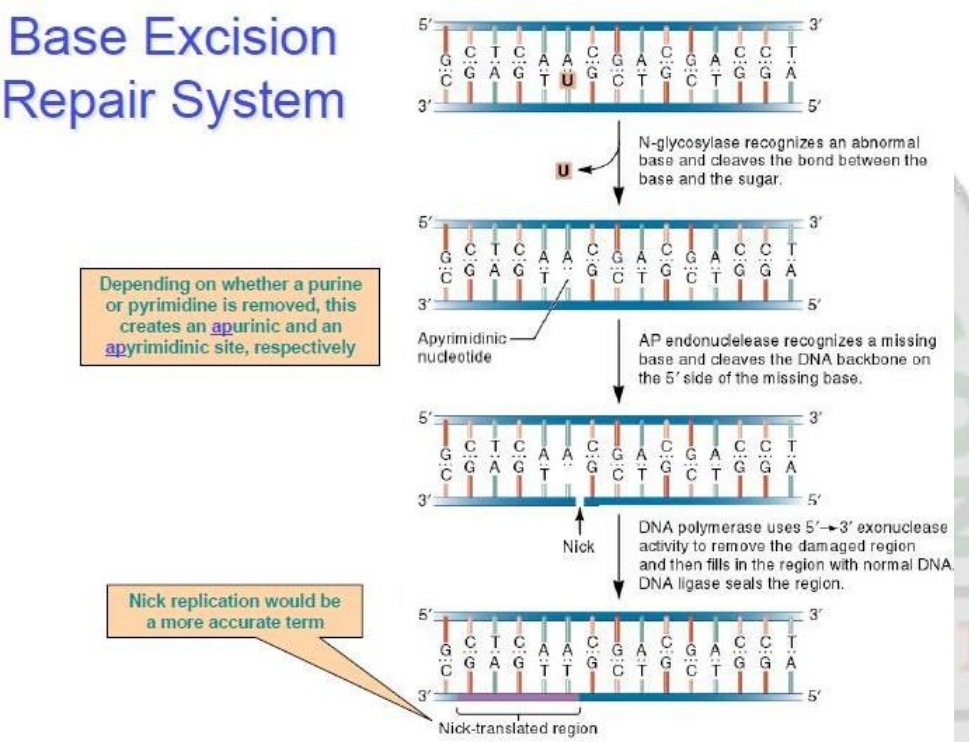
DNA repair:

The mis-incorporation or damages of DNA during replication is rectified by DNA repair system. Following are the types of DNA repair:

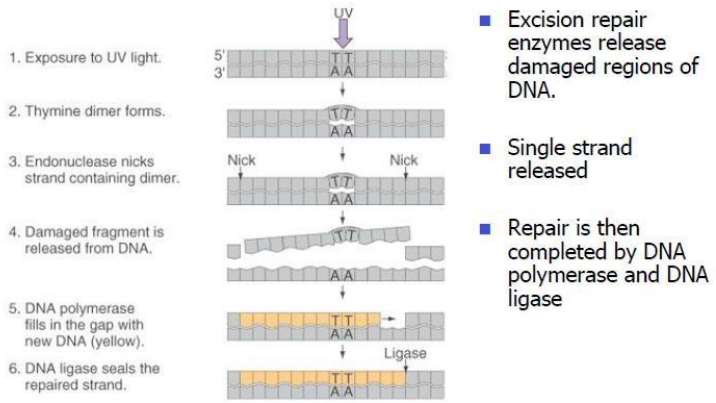
A. Direct repair: In a few cases, the covalent modifications of nucleotides can be reversed by specific enzymes. For example, Photolyase can repair thymine a repair thymine dimers induced by UV light nduced by UV light. It splits the dimers restoring the DNA to its original condition. In a similar way, O^6-alkylguanine alkyltransferase repairs alkylated bases. It transfers the methyl or ethyl group from the base to a cysteine side chain within the alkyltransferase protein.



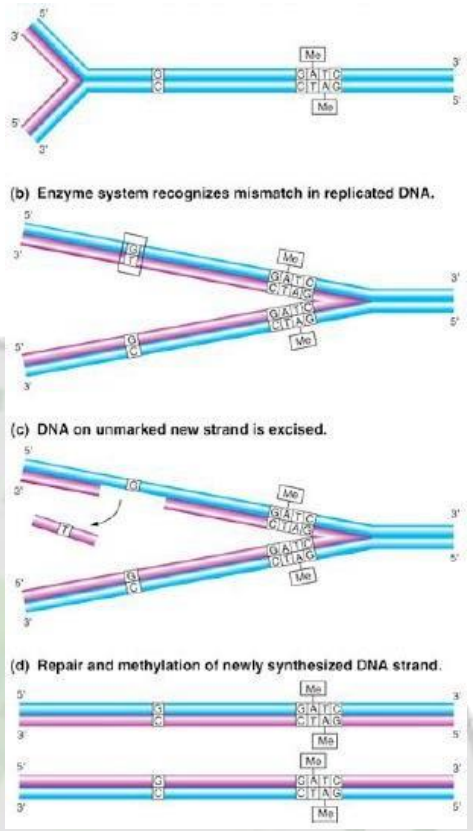
B. Base excision repair: It involves a category of enzymes involves a category of enzymes known as DNA-N-glycosylases. These enzymes can recognize a single damaged base and cleave the bond between it and the sugar in the DNA. It removes one base, excises several around it, and replaces with several new bases using Pol adding to 3‟ ends then ligase attaching to 5‟ end. Depending on the species, this repair system can eliminate abnormal bases such as-Uracil: Thymine dimers, 3-methyladenine: 7-methylguanine.



C. Nucleotide Excision Repair: It nicks DNA around damaged base and removes region, and then fills in with Pol on 3‟ends, and attaches 5‟ end with ligase. This type of system can repair many types of DNA damage, This type of system can repair many types of DNA damage, including Thymine dimers and chemically modified bases. This type of repair system is found both in prokaryotes and eukaryotes.



D. Mismatch repair: If proofreading fails, the methyl-directed mismatch repair system comes to the rescue. This repair system is found in all species. In humans, mutations in the system are associated with particular types of cancer. Methyl-directed mismatch repair recognizes mismatched base pairs, excises the incorrect bases, and then carries out repair synthesis



CHAPTER 14

TRANSCRIPTION AND TRANSLATION

I. Transcription (General info)

A. Transcription is the synthesis of RNA using DNA as a template.

B. Early evidence suggesting an RNA intermediate between DNA and proteins

1. DNA was in the nucleus but proteins were made in the cytoplasm

2. RNA synthesis in the nucleus was exported to the cytoplasm

3. T2 infection of *E. coli* results in phage specific RNA being produced

C. Properties of RNA – Similar to DNA except

1. Contains ribose instead of deoxyribose

2. Contains uracil instead of thymine

3. Single stranded instead of double stranded (although there are regions of pairing)

D. Misc other info:

1. Each RNA species is complementary to one strand (template strand) of the DNA double helix.

2. Upstream vs. downstream: RNA strand has a 5‟ and 3‟ end. Upstream refers to “towards the 5‟ end” and downstream refers to “towards the 3‟ end”.

3. The region of DNA that contains sequences that are the signals for transcribing a gene are termed promoters.

4. +1 refers to the basepair where transcription starts; -x refers to x basepairs 5‟ to the start site

II. Factors required for transcription

A. Prokaryotic

1. RNA polymerase (enzyme that catalyzes the synthesis of RNA from a DNA template).

a) Core enzyme = 3 different types of subunits (2a; 1b; 1b‟)

(1) b - binds incoming nucleotides

(2) b‟ – binds DNA

(3) a - helps with enzyme assembly; interacts with other transcriptional activator proteins; recent work demonstrated that a also interacts with some DNA sequences

b) Holoenzyme = core + s factor (recognizes the promoter)

c) s factors – Initially, people thought that there was only one s factor that functioned to direct RNAP to the promoters of genes. Later, different classes of s factors were found. Each s

factor directs RNAP to a different type of promoter (differentiated by a specific DNA sequence in the promoter).

2. Accessory transcription activator proteins

a) Can bind to specific DNA sequences and help RNA polymerase initiate transcription via protein-protein interactions or by altering the structure of the DNA.

b) Transcription of some promoters requires an accessory transcriptional activator; at other promoters, the activators just increase the rate of transcription but are not absolutely required.

3. Template DNA containing gene or genes to be transcribed

4. Promoter - The regulatory element that determine when a gene "turned on" (transcribed) or "turned off". The promoter DNA is located upstream of the gene and contains a sequence which s factor of RNAP and other transcription factors bind. Different classes of promoters have different DNA sequences. Deviations from the consensus sequence decrease the level of transcription.

5. Weak promoters (ones that have poor sigma recognition sequences) have additional sequences to which transcriptional activators can bind. 6.NTPs, Mg 2+

B. Eukaryotic 1.RNA polymerases – Much more complex that prokaryotic RNAP (numerous additional factors required, multiple polymerases) a)RNAP I –synthesizes ribosomal RNA b)RNAP II –synthesizes messenger RNA c)RNAP III – synthesizes transfer RNA and 1 type of rRNA

2. Eukaryotic RNAPs have subunits that are homologous to α,β, and β‟of prokaryotic RNAP; however, eukaryotic RNAP also contain many additional subunits.

3. Template DNA containing the gene to be transcribed 4.Eukaryotic promoters – contain some combination of the following a)contain a TATA rich region located –25 to -30 from the start of transcription b) Upstream from the TATA region is a variably located sequence containing the sequence CCAAT (frequently at –75) c)GC box d)Some promoters have other sequences located either upstream or downstream that maximize the level of transcription called enhancers 5.NTPs, Mg 2+

III. Prokaryotic transcription

A.Initiation 1.RNAP scans the DNA looking for promoters. 2.□factor of RNAP binds the corresponding □ factor recognition sequence in the promoter. 3.Recent evidence suggests that at some promoters, the □ subunit may bind to AT rich regions upstream of the sigma

binding sites. 4.RNAP is bound covering approx. 60 basepairs. The DNA is still is a double helix (closed complex). 5.RNAP unwinds the DNA resulting in open complex formation. 6.First nucleotides are added to start RNA chain. Transcriptional initiation has occurred! 7.Accessory transcription factors may aid in all of the above listed steps.

B.Elongation

1. Elongation is 5‟ 3‟

2. σfactor is ejected from RNAP after first 2-10 nucleotides are added.

3. Much less is known about this step for transcription than initiation. It was once believed that elongation occurred at a constant rate; however, recent work suggests that RNAP may pause during elongation. In fact, pausing is important in termination

C. Termination (2 types)

1. Rho independent: A specific sequence at the end of the gene signals termination. The sequence is transcribed into RNA and it is the RNA sequence that is important. This sequence contains numerous Gs and Cs, which forms a hairpin structure, followed by a string of Us. The hairpin destabilizes the DNA:RNA hybrid leading to dissociation of the RNA from the DNA.

2. Rho dependent: Rho protein binds to a sequence in the RNA (rut site – not well characterized). Rho moves along the RNA in the 3‟direction until in eventually unwinds the DNA:RNA hybrid in the active site, thereby pulling the RNA away from the DNA and RNAP. Rut sites are located 5‟to sites in the DNA that cause RNAP to pause. It is thought that this allows Rho to catch up to RNAP and the RNA-DNA hybrid.

IV. Eukaryotic transcription

A. Initiation and elongation are similar to in prokaryotes; however, there are several important differences.

Prokaryotes	Eukaryotes
1. All RNA species are synthesized by a single RNA polymerase.	1. Three different RNA polymerases are responsible for the different classes of RNA molecules.
2. mRNA is translated during transcription.	2. mRNA is processed before transport to the cytoplasm, where it is translated. Caps and tails are added, and internal portions of the transcript are removed.
3. Genes are contiguous segments of DNA that are colinear with the mRNA that is translated into a protein.	3. Genes are often split. They are not contiguous segments of coding sequences; rather, the coding sequences are interrupted by intervening sequences (introns).
4. mRNAs are often polycistronic.	4. mRNAs are monocistronic.

B. Termination of transcription in eukaryotes is poorly understood.

C. RNA processing

1. 5‟capping: Occurs early in transcription.Guanosyltransferase adds 5‟ methyguanosine (Cap) to 5‟end of mRNA. The Cap is important for translation initiation and for export from the nucleus.

2. 3‟poly(A) tail: AAUAAA sequence in the RNA signals a cleavage event in the RNA.Poly(A) polymerase then adds 150-200 A residues are added to the 3‟ end of the mRNA. The poly(A) tail increases the stability of the mRNA in eukaryotes.

Recent evidence has demonstrated that there are poly(A) polymerases in prokaryotes and that some mRNAs have poly(A) tails. Interestingly though, the polyA tail destabilizes the mRNA in prokaryotes. Someα2-thalassemias (anemia due to imbalance of α and β hemoglobin subunits) have been attributed to a defect in polyadenylation. Specifically, there is a mutation in the cleavage site from AAUAAA AAUAAG.

3. Splicing: The primary transcripts often contain intervening sequences (introns) that are removed from the RNA prior to translation by a cleavage reaction catalyzed by snRNPs (small nuclear ribonuclear proteins which contain RNA and protein). Frequently, the splicing site in the intron has a GU at the 5‟ end and an AG at the 3‟end. The snRNP aligns these ends in a lariat formation to allow precise splicing.

Complexes containing the snRNP, mRNA, and associated proteins are called spliceosomes.

Splicing is important (1) splicing allows variations of a gene and therefore gene product to be made (2) it has been suggested that exons correspond to functional motifs in proteins and thus the presence of genes that require slicing allows for evolutionary tinkering (3) many viruses have spliced mRNAs and so understanding the process may lead to new therapeutic approaches.

As an interesting aside, people with systemic lupus erythematosus have antibodies directed against snRNP protein subunits. The significance of this is unknown at this time.

D. RNA export: RNA synthesis and processing occurs in the nucleus. The mature mRNA is then transported through the nuclear pores in the nuclear envelope to the cytoplasm. There is a nuclear complex that is involved in the transport. This complex recognizes the 5‟CAP of the mRNA.

V. Translation –

General info

A. Translation is the production of a polypeptide (protein) using RNA as a template and tRNA molecules as “adapters”that convert the nucleic acid code to protein code.

B. The nucleotides (letters) of RNA formed codons (words) that specify a particular amino acid.

C. The tRNA contains an anticodon that is complementary to the codon and carries a specific amino acid.

D. Important elements of the genetic code: 1.The code is a triplet code: Each mRNA codon (word) that specifies a particular amino acid in a polypeptide chain consists of three nucleotides (letters). For example, AAG = lysine 2.The code is non-overlapping: The mRNA encoding one protein is read in successive groups of three nucleotides. 3. The code is degenerate: More than one mRNA codon (word) occurs for some amino acids (ie. AAG and AAA are read as both read as lysine)

a) Wobble – certain different codons are recognized by the same tRNAs because the 3 rd base in the codon and the 1 st base of the anticodon pair via a“loose pairing”. This “loose pairing is according to a set of rules known as the wobble rules.

b) There is more than one tRNA type (therefore more than one anticodon) for some amino acids.

4. The code has start signals (AUG and rarely GUG) and stop signals (UAA, UAG, and UGA). Stop signals are also called nonsense codons because they do not designate an amino acid.

5. The code is commaless.

6. The code is almost universal.

VI. Factors required for translation

A. Prokaryotic

1. mRNA – contains a RBS (ribosome binding site) / also known as a Shine-Delgarno sequence. The RBS is characterized by a core sequence 5‟AGGAGU3‟ located 7+2 nucleotides from the AUG. Deviations from the consensus decrease translation.

2. tRNA – adapter molecules in the information transfer between mRNA and protein which has: a)anticodon which is a 3 nucleotide sequence that is complementary and antiparallel to the mRNA codon b)amino acid attachment site at the 3‟end for attachment of the amino acid c)3-D shape that determines which amino acid will be attached to the amino acid attachment site. Recent studies indicate that the anticodon loop, the D loop, and the aminoacyl stem are all important. The correct attachment of the amino acid to its tRNA is considered the “2 nd genetic code”and is still being cracked. d)Isoacepters = tRNAs with different anticodons but same amino acid. e)Aminoacyl tRNA = tRNA with amino acid attached = charged tRNA.

3. there are 20 of these, each recognizing 1 amino acid and all the tRNAs that to which that amino acid is to be attached

4. Ribosomes (Note *S* refers to a sedimentation value of the structure in a sucrose gradient) a)Large subunit (50*S*)– consists of 23*S* and 5*S* rRNAs and 31 ribosomal proteins b)Small subunit (30*S*)- consists of 16*S* rRNA and 21 ribosomal proteins

5. Soluble transcription factors a)Initiation factors (1)IF1 – promotes dissociation of ribosomal subunits (2)IF2(•GTP) – required for fMET-tRNA met binding (3)IF3 – required for mRNA binding, finding the AUG b)Elongation factors (1)EF-Tu (•GTP) – binds aminoacid-tRNA to the ribosome (2)EF-Ts – regenerates EF-Tu•GTP (3)EF-G(•GTP) – increases translocation rate c)Termination factors (1)RF1 – recognizes UAA and UAG stop codons (2)RF2 – recognizes UAA and UGA nonsense codons (3)RF3(•GTP) – enhanced RF-1 and –2 binding to ribosome

6. Amino acids

7. F-met (N-formyl Met-tRNA)

8. GTP

9. ATP (for charging tRNAs)

B. Eukaryotic (similar to prokaryotes except…)

1. One gene per mRNA (monocistronic)

2. Although the processes are similar, the component of eukaryotic and prokaryotic translation can not be mixed

3. Ribosomes interestingly, only two ribosomal proteins and the rRNA which are very highly conserved among prokaryotes and eukaryotes. For euks, a)Large subunit (60*S*)– consists of 28*S*, 5.8*S*, and 5*S* rRNAs and 50 ribosomal proteins b)Small subunit (40*S*)- consists of 18*S* rRNA and 33 ribosomal proteins 4.Soluble translation factors a)Initiation factors (1)eIF1 – promotes dissociation of ribosomal subunits (2)eIF2(GTP) – required for fMET-tRNA met binding (3)eIF3 – (4)eIF4 – important for finding the capped end of the mRNA b)Elongation factors (1)EF (•GTP) – binds AA-tRNA to the ribosome (2)EF 1β – regenerates EF •GTP (3)EF 2 (•GTP) – increases translocation rate c)Termination - Several TF (termination factors) 5.No F-met

VII. Translation – Mechanism in Prokaryotes

A. Initiation – the purpose of this step is to set the reading frame 1.IF1, IF2•GTP, and IF3 bind to the 30*S* subunit. 2.Binding of mRNA to the 30*S* subunit via an interaction between the RBS on the RNA and a complementary sequence at the 3‟end of the 16*S* RNA. Facilitated by IF3. 3.Release of IF3 4.fMET•tRNA binds to the P site in the 30*S* subunit with the help of IF-2. 5.GTP hydrolysis and release of IF1 and IF2 drives the attachment of the 50*S* subunit.

B. Elongation – addition of amino acids to the growing polypeptide chain 1.EF-Tu•GTP-AA-tRNA complex binds to the A site (there is a selection process that goes on at this step whereby if the match between the codon and anticodon is not correct, the complex is released before the next step can occur) 2.GTP hydrolysis 3.Proofreading (if the match between the codon and anticodon is not correct, the complex is released before the next step can occur) 4.EF-Tu release (Note that EF-Tu•GTP is regenerated via the action of EF-Ts) 5.Peptidyl transfer – polypeptide is transferred from the tRNA at the P site to the AA-tRNA complex at the A site. This catalytic activity is thought to involve not only proteins but also the 23*S* RNA. 6.Translocation – shift of the ribosome one codon towards the 3‟end resulting in transfer of the tRNA with the polypeptide chain to the P site (stimulated by EF-G). During translocation the uncharged tRNA in the P site is moved to the E site (for exit) which is thought to block the A site unit translocation is complete.

C. Termination a)A termination codon (UAA, UAG, UGA) is presented at the A site b)RF1 or RF2 bind to the A site with the help of RF3•GTP c)The RF-mRNA-ribosome complex catalyzes peptidyl hydrolysis instead of transfer d)The polypeptide is released from the tRNA in the P site. e)The GTP associated with RF3 is hydrolyzed causing the release of the 3 RF factors and the tRNA from the ribosome f)The 30*S* and 50*S* subunits dissociate with the aid of IF1 and IF3 and the mRNA is released

VIII. Importance in understanding translation in detail.

A. Translation is a fundamental process to all life.

B. Antimicrobial drug design against components of translation machinery that are different between eukaryotes and prokaryotes.

C. Antisense DNA designed to bind to the beginning of specific mRNAs to prevent transcription.

CHAPTER 15

THE *Lac* OPERON AND FINE STRUCTURE OF GENE

Specific proteins are present in different tissues and some appear only at certain times during development. All cells of a higher organism have the full set of genes.

Regulation can occur at all levels:

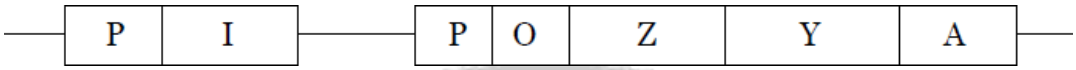
multiple genes
promoter efficiency
mRNA stability
translation
post translational modification
protein stability,

Regulation of transcription: is especially effective because mRNA typically has a short half life (1.8 minutes in *E. coli*) so stopping mRNA synthesis leads to rapid changes in protein synthesis. it takes lots of energy to make mRNAs (and proteins) so making them when they are not needed is inefficient.

The Lac Operon has to do with the ability of *E. coli* to utilize the sugar lactose. Lactose is a 12 Carbon sugar made of 2 simpler 6 carbon sugars, glucose and galactose. Glucose is a very efficient carbon source; it can enter directly into the metabolic paths that provide both energy and substrates for making more complex compounds. If lactose is provided as the carbon source, it must first be broken down into the two component sugars before it can be used. The enzyme for breaking down lactose in *E. coli* is called β-galactosidase. The following observations demonstrated that the gene that codes for β-galactosidase in *E. coli* is regulated: *E. coli* grown in glucose as the sole carbon source have about 3 copies of the enzyme β-galactosidase/cell. *E. coli* grown in lactose as the sole carbon source have about 3,000 copies of the enzyme β-galactosidase/cell. The system of regulation seen here is called "induction" since synthesis of the enzyme is "turned on" only when needed. Induction typically is used to regulate "breakdown" (catabolic) pathways as opposed to "synthetic" (anobolic) pathways. Francios Jacob and Jaque Monod won a Nobel prize for their work in describing how the lac operon functions. They used a genetic approach to address the problem, by identifying mutants that did not have normal regulation of β-galactosidase. We will first look at the model they derived, and then see how the behavior of mutants led to the model.

The lac-operon is actually a series of adjacent genes and regulatory elements in one small part of the *E. coli* circular chromosome.

Lac-Operon components



Definitions:

P strands for promoter; it is the site where RNA polymerase attaches in order to transcribe mRNA. Although all promoters have the same function and share similar sequences that are recognized by RNA polymerase, they differ enough so that some are very strong (leading to high levels of transcription) and others are weak (rarely transcribed). Thus, one level of regulating gene expression comes as a consequence of the strength of the promoter at the beginning of the gene.

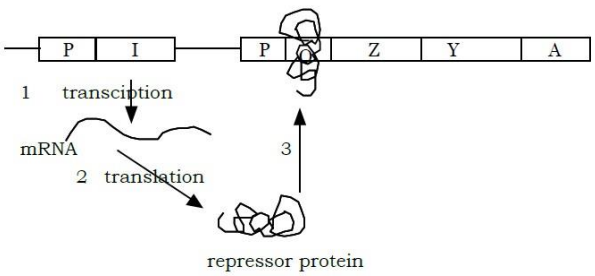
The *I* gene is called a regulator gene; it is transcribed to make a mRNA which is translated to a repressor protein. There is a termination signal at the end of the *I* gene.

O stands for Operator; it is a short sequence of bases that acts like a switch that can be recognized by repressor protein.

Z, *Y* and *A* are all "structural genes (genes that code for polypeptides) Z codes for β-galactosidase; Y codes for lactose permease, a protein that functions to actively bring lactose from outside to cell to the inside, even against a concentration gradient. A codes for transacetylase, an enzyme that is also needed to breakdown many sugars related to lactose.

One long mRNA is made for the Z, Y and A genes; this is the basis for the system being called an operon. All 3 genes that code for enzymes needed to use β-galactoside molecules as a source of carbon and energy are adjacent and are coordinately turned on or off by regulating transcription. Operons are only found in prokaryotes; in eukaryotes, each structural gene has its own promoter and regulatory elements.

Lac-operon function when only glucose is present; that is when we expect it to be turned off (numbers indicate steps in the description):



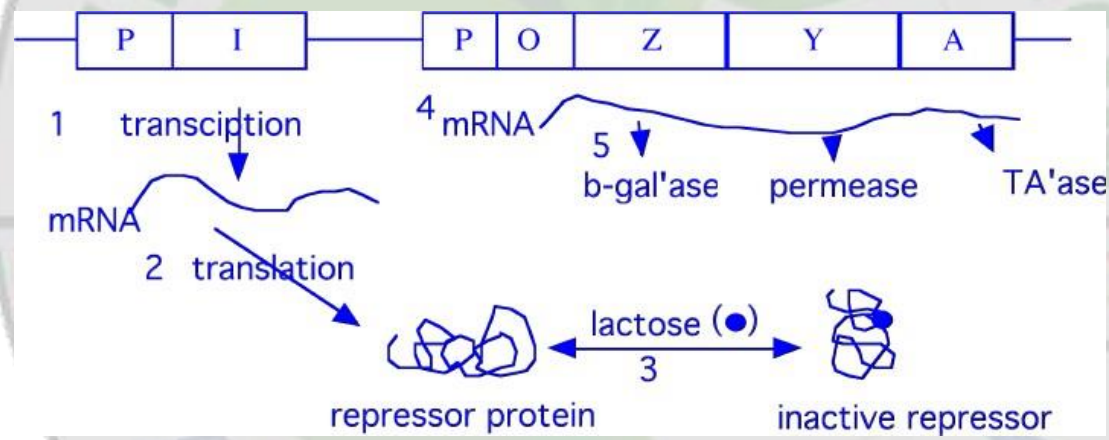
Stepwise:

1. The Promoter for the I gene is always "on", but is very weak, so it is transcribed only rarely. A gene that is not regulated, other than by the strength of its promoter, is said to be "constitutive".

2. The I mRNA is translated into a polypeptide; 4 copies make one repressor protein. A typical cell will have only about 10 copies of this protein.

3. In the absence of lactose, the repressor protein binds to the operator, preventing transcription from the second promoter. Almost no ZYA mRNA is made. (The operator is split in 2 parts each with 28 of 35 bases in a palindrome; when the repressor binds it "folds" the DNA so that the promoter is not accessible).

When only lactose is present the model works as follows:



Stepwise:

1. The Promoter for the I gene occasionally is bound by an RNA polymerase to initiate transcription.

2. The I mRNA is translated into the repressor protein.

3. Lactose (actually one stereo-isomer called allolactose which is a minor product of β−gal'ase function) binds to the repressor very efficiently and converts the repressor into an inactive state, where it can't bind the Operator. The process is reversed when all the lactose is digested, so the system again will turn off.

4. When the very strong Promoter for making Z-Y-A mRNA is not blocked, many copies of the mRNA are made. The small amount of lactose that diffuses in is able to initiate induction of transcription of the Z-Y-A mRNA. Even as the message is being made, translation begins and the 3 proteins are made.

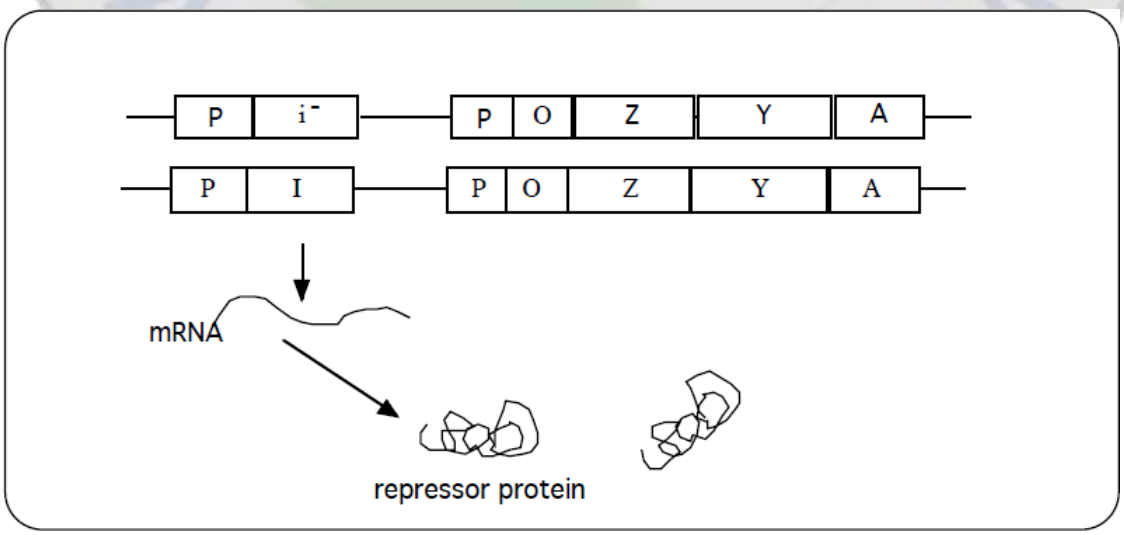
5. Translation begins at the 5' end of the mRNA and makes β-galactosidase from the Z gene. There is a stop codon, followed immediately by another AUG start, so many, but not all, ribosomes read on through and make permease from the Y gene. The same process allows some A gene product to also be made.

Mutations that define the Lac-Operon model:

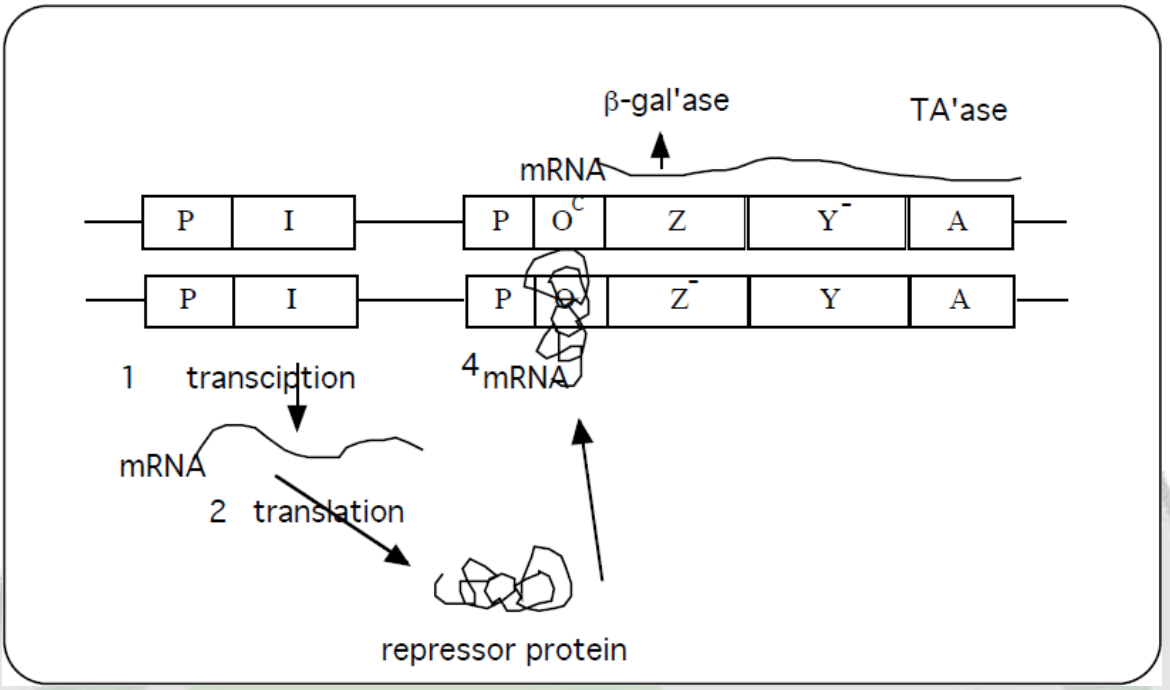
Jacob and Monod found mutants that did not show the normal regulation, that is, synthesis of Z, Y, and A proteins only when lactose was present.

Two kinds of mutants gave constitutive (continuous) synthesis of β-galactosidase, permease and TAase, no matter what carbon source was present

a) Mutants in the I gene (i- or ic) mutants all were mapped (we will take up mapping later in the course) to a similar location. F'lac strains were used to make recipient cells that were partial diploids with two copies of the lac-operon. When the copies created heterozyous cells (I+, I-) normal regulation was observed. This indicated that the I gene codes for something that can move and interact with the operators of both copies of the lac-operon present in these cells.



b) Mutants in the promoter that change the base sequence so that it is no longer recognized by the repressor protein are also constitutive for Z, Y and A expression. In this case however, the mutation is dominant in partial diploids:



Since the operator in the upper strand cannot be bound by active repressor, β-galactosidase and TA'ase will always be made. In this cell, permease will show normal regulation since it is only made by the lower copy of the lac-operon. These results told Jacob and Monod that the Operator regulated transcription only of gene on the same DNA molecule.

Mutations in the I gene that prevent the repressor protein from interacting with lactose, but still bind to the operator and permanently turn off all transcription. These are called iS mutations for superrepressors.

The partial diploids told Jacob and Monod that the I gene coded for something that could move about the cell and interact with any DNA. Now such gene products are referred to as "trans-acting factors". iS mutations are an example of a trans-dominant effect.

The operator mutations only prevented transcription from the same DNA strand; thus they are "cis-elements" and in this case, act as cis-dominant mutations.

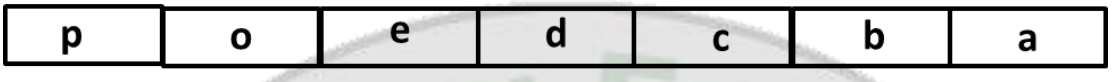
Some technical details:

If both glucose and lactose are present, cells use up the glucose before turning on the *lac* operon. When energy begins to become limiting, a signal molecule (cAMP = cyclic-AMP) builds up, binds to a catobolite activating protein (CAP); and the complex in turn binds to a site between the promoter segments of the lac operon. Binding of the cAMP/CAP complex opens the promoter for RNA polymerase binding (providing the lac-repressor isn't bound also).

CHAPTER 16

THE *TRP* OPERON

The trypotophan (*trp*) operon n in *E. coli* controls the enzymes that catalyse the biosynthesis of the amino acid tryptophan, where 5 gens are involved. The *trp* operon is an example of repressible operon.



The gene functions are as follows:

trpE+trpD: Anthranilate synthetase

trpC: Indole glycerolphosphate synthetase

trpB+trpA: Tryptophan synthetase

p: Promoter

o: Operator

There is a separate gene (r), that encodes the repressor protein.

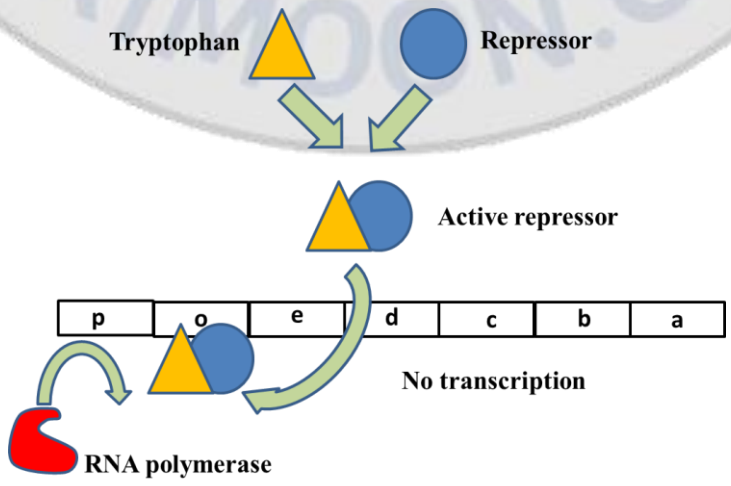
The regulation of the expression of the genes is governed mainly by 2 mechanisms:

a. Control by the repressor

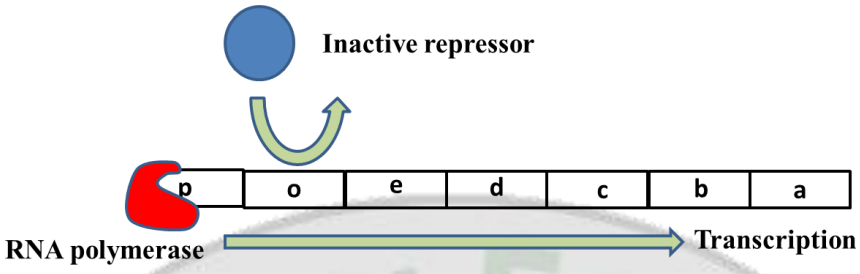
b. Control through attenuation

a. Control by the repressor:

When tryptophan is present in sufficient amount in the cell, it acts as a corepressor. Binding of the corepressor with the repressor changes the conformation of the repressor and allows the repressor/corepressor complex to bind in the operator DNA, changes conformation and thus prevents transcription by inhibiting the binding of the RNA polymerase to the promoter region.

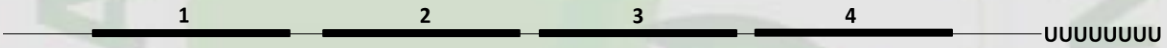


When sufficient tryptophan is absent in the cell, the repressor remains inactive and cannot bind to the operator region. This allows RNA polymerase to transcribe the genes of the trp operon for tryptophan biosynthesis.

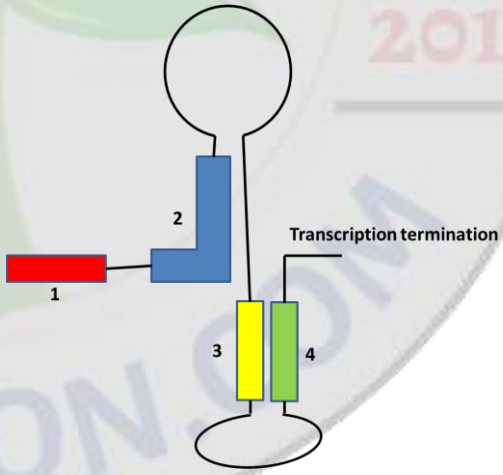


b. Control through attenuation

Transcriptional attenuation is a second, separate control mechanism. There is an attenuator region between the operator DNA and structural genes (*edcba*) in the *trp* operon. RNA from the attenuator region is called the leader transcript. This contains 4 regions (1,2,3 and 4) than may form stem-loop structures, with three possible pairings: 1-2, 3-4, and 2-3. The stem-loop 3-4 is followed by 8 U‟s: which acts as a typical ρ-independent transcription termination signal. The attenuator region also encodes a leader peptide – 14 aa‟s, with two adjacent trp codons.



When tryptophan is high, the leader RNA is transcribed, ribosome binds and translates leader peptide covering regions 1 & 2, and then 3 & 4. Transcription terminates via ρ-independent mechanism due to termination stem-loop formed between 3 & 4 and adjacent U‟s . Structural genes are not transcribed due to early termination (attenuation).



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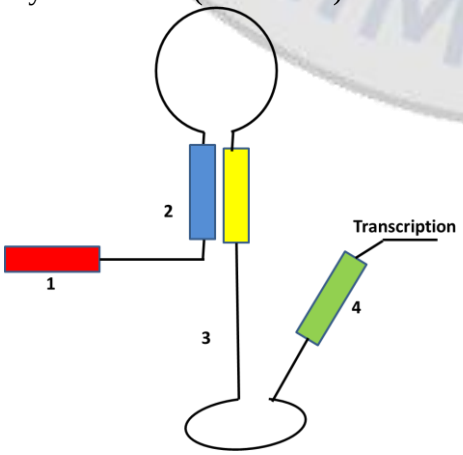


When tryptophan is low, the leader RNA is transcribed, but the ribosome is stalled at the trp codons in leader peptide due to absence of sufficient trp-tRNA. Region 2/3 stemloop is formed, preventing the formation of the 3/4 transcription termination stem-loop. This allows expression of the trp operon structural genes.

CHAPTER 17

EVOLUTION OF CROP SPECIES AND CHROMOSOMAL ABERRATIONS

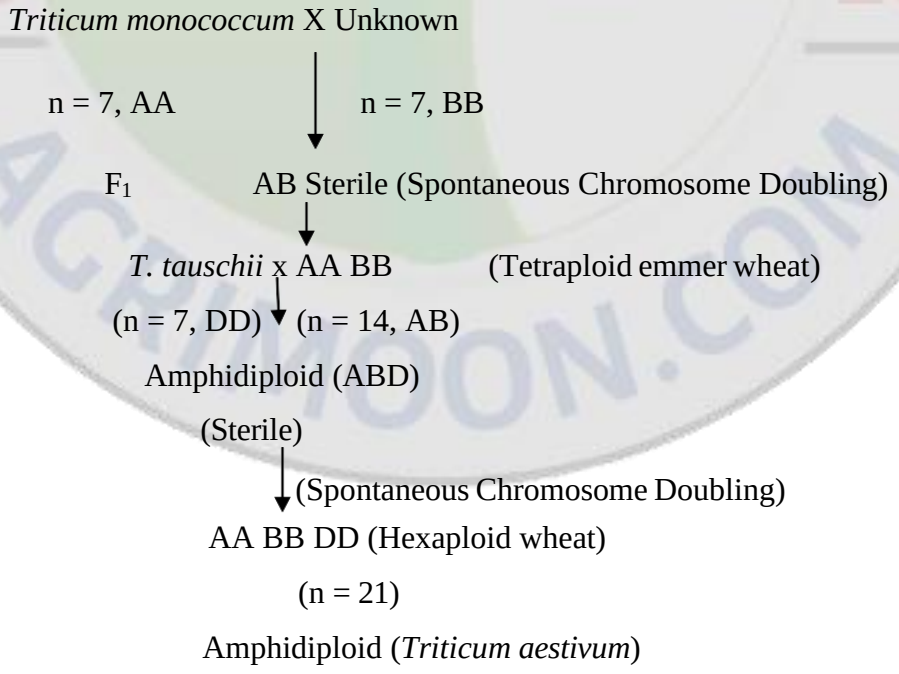
Role of Allopolyploidy in Evolution:

It is estimated that about one-third of the Angiosperms are polyploids, and by far the vast majority of them are allopolyploids.

The identification of parental diploid species is primarily based on pairing between the chromosomes of the diploid and the allopolyploid species. When the chromosomes of a diploid species pair with some of those of the allopolyploid species, homology between chromosomes of the two species is apparent. This homology suggests that the diploid species may be one of the parental species of the allopolyploid.

We shall briefly consider the possible evolutionary history of some important allopolyploid crop species, *viz.*, wheat, tobacco, cotton and Brassica.

Evolution of Bread Wheat (*Triticum aestivum*). Evolutionary history of wheat has been the most extensively investigated, and is perhaps the least understood. Identity of the diploid species contributing the three genomes (A, B, and D genomes) of *T. aestivum* has been investigated by many workers, more notably by Sears, Kihara and others.



Evolution of Nicotiana tabacum. *N. tabacum* (n = 24) is most likely an amphidiploid from the cross *N. sylvestris* x *N. tomentosa*; both the species are diploid with n = 12. The

interspecific hybrids *N. tabacum* x *N. sylvestris* and *N. tabacum* x *N. tomentosa* produce 12 II and 12 I at metaphase I. This indicates a homology between chromosomes of *N. tabacum* and those of *N. sylvestris* and *N. tomentosa*. The amphidiploid from the cross *N. sylvestris* x *N. tomentosa* is similar to *N. tabacum* in many characteristics, which further supports the above conclusion. The species *N. tabacum* has undergone considerable differentiation during its evolutionary history, mostly due to the accumulation of gene mutations and, to some extent, due to the loss of some duplicated segments of the two genomes.

Evolution of *Gossypium hirsutum*. The 9 old World and the 8 New World species of Gossypium have n = 13, but the chromosomes of the New World species are smaller than those of the Old World species. Three other species, *G. hirsutum*, G. barbadense and *G. tomentosum* (wild Hawaii cotton), have n = 26; in these species, 13 chromosomes are relatively larger than the remaining 13. A possible origin of *G. hirsutum* is from the cross between Asiatic cotton *G. arboreum* x *G. thurberi* (American wild cotton), followed by chromosome doubling of the inter specific hybrid. According to a more recent scheme, *G. hirsutum* has originated from the cross *G. herbaceum* var. *africanum* x *G. raimondii*, followed by chromosome doubling of the F_1.

Evolution of Amphidiploid *Brassica* Species*:* The origin of amphidiploid *Brassica* species is presented based on the famous U‟s Triangle proposed by N. U in 1935. According to this scheme, *B. juncea* (n = 18) is an amphidiploid from *B. nigra* (n =8) x *B. campestris* (n = 10), *B. napus* (*n* = 19) is an amphidiploid from the cross *B. oleracea* (*n* = 9) x *B. campestris* (n = 10), and *B. carinata* (n = 17) is an amphidiploid from the cross *B. nigra* (n = 8) x *B. oleracea* (n = 9). The synthetic allopolyploids produced according to the above scheme resemble the natural amphidiploids, cross easily with them, and the hybrids between the synthetic and natural amphidiploids are reasonably fertile.

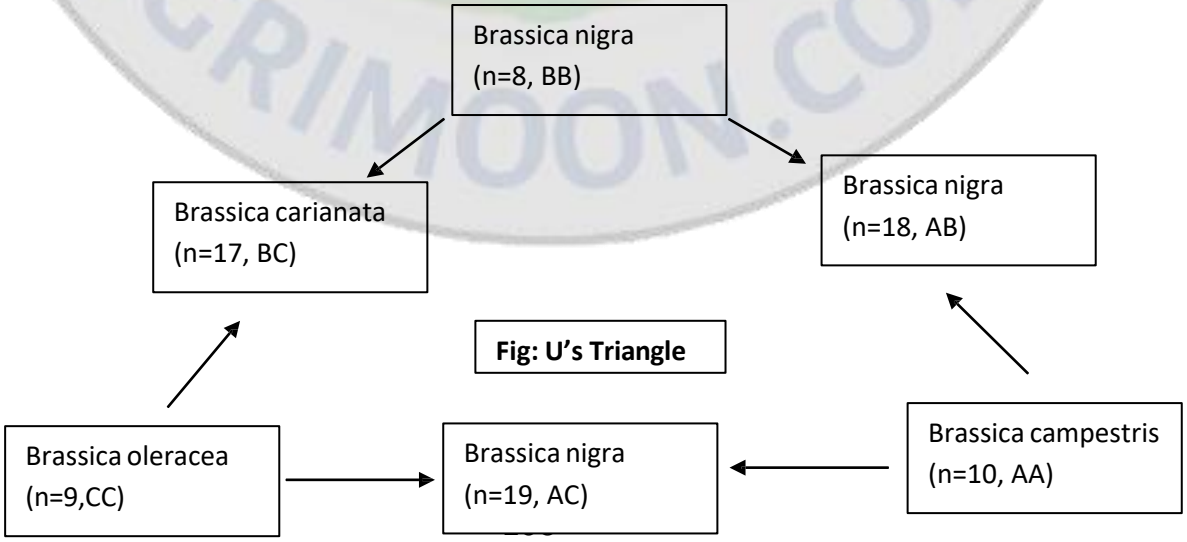


Fig: U's Triangle

Numerical Chromosome aberration:

Aneuploidy:

Of the various anecuploids, monosomics (in polyploid species, such as, tobacco, wheat and oats) and trisomics [in diploid species, *e.g.*, maize, bajra, tomato, rye, pea, spinach, etc.] are most commonly used in genetic studies.

Nullisomics are viable in a few highly polyploid species only, *e.g.*, wheat and oats; they are not viable even in tobacco. Aneuploids are usually less vigorous than their diploid progenitors. Another characteristic of aneuploids is their high sterility resulting form irregular meiosis.

Monosomics

A monosomic is an individual that lacks one chromosome of the normal complement of somatic cells (2n – 1).

If the lost chromosome is one that is not absolutely essential for the organism, it may survive but if the lost chromosome is one that is very important, it may not live.

Loss of one chromosome in normal diploid plants may result in lethality. Thus, for example, monosomics are inviable in *Datura sp.* Polyploid plants, however, have been found to tolerate the loss of one chromosome. Twenty-four different monosomics, each lacking a single different chromosome of the normal complement, have been isolated in *Nicotiana tabacum* which is a tetraploid with 2n = 48. These 24 monosomics are morphologically distinct from each in haploid wheat (2n = 42), 21 different monosomics have been isolated.

Monosomics produce two kinds of gametes, one kind with n chromosomes and the other kind with n –1 chromosomes. When selfed, monosomics, therefore, produce normal (i.e., disomic), monosomic and nullisomic offspring.

Nullisomics

A nullisomic is an individual that lacks both members of one specific pair of chromosomes (2n-2).

Nullisomics are inviable in some species like *Nicotiana tabacum*, but in other species like *Triticum aestivum*, they are viable. In the Chinese Spring variety of wheat, Sears established 21 nullisomic lines (2n = 40), each lacking a single pair of chromosomes of the normal

complement of the somatic cells. Different nullisomics are morphologically different from one another and from the normal Chinese Spring. They are reduced in size and vigour and are highly sterile. On selfing, they produce only nullisomics as their gametes contain only n – 1 (i..e, 20) chromosomes each.

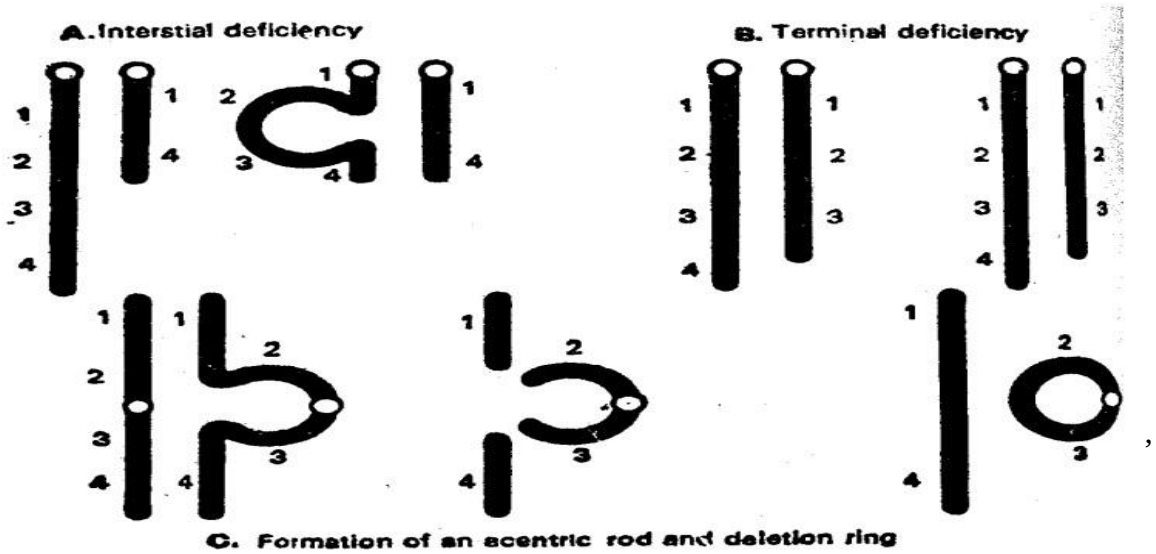
Structural chromosomal aberrations

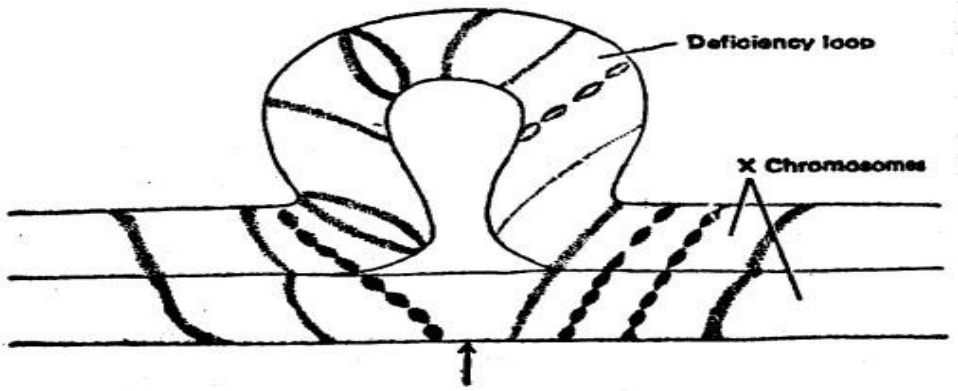
Types of chromosomal aberrations: The chromosomal aberrations may remain confined to a single chromosome or may extend to both of the member of the homologues pair and, therefore, may be of following types:

A. Intra chromosomal aberrations B. Inter chromosomal aberrations.

A. Intra chromosomal Aberrations When aberrations remain confined to a single chromosome of a homologous pair, they are called intra chromosomal or

1. Deficiencies (Deletions) In deletion or deficiency type intra chormosomal aberration a chromosomal lacks either in an interstitial or terminal chromosomal segment which may include only a single gene or part of a gene. If break occurs near the end of a chromosome, a small piece of the terminal end is lost and thus, terminal deficiency occurs. Sometimes, two breaks may occur at any two points, releasing an intercalary segment which may remain rod-shaped or may become ring shaped, if its broken ends join and fuse. If, this ring-shaped chromosome (called deletion ring) has centromere is persists, but if lacks in that, loses during cell division. The broken ends of original chromosome are fused and has intercalary or intersitial deficiency. If this chromosome has centromere it persists otherwise lost during cell division.



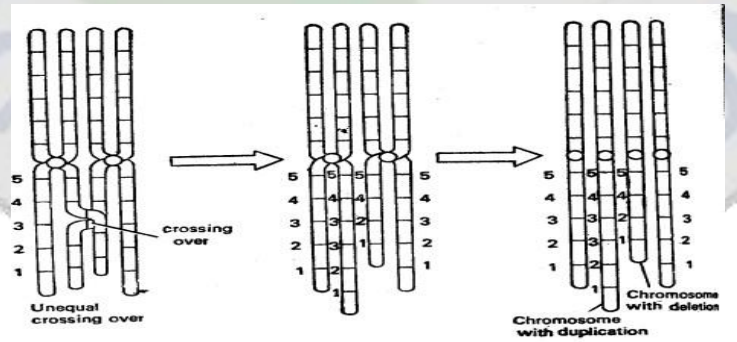


A deficiency loop in the paired X-chromosomes from a salivary gland cell of a *Drosophila* larva heterozygous for notch (after King, 1965).

Genetic Significance of Deficiencies Lethal effect: Organisms with homozygous deficiency usually do not survive to an adult stage because a complete set of genes is lacking.

2. Duplications (Additions)

Duplication occurs when a segment of the chromosome is represented two or more times in a chromosome of a homologous pair. This extra-chromosomal segment may be a free fragment with a centromere or a chromosomal segment of the normal complement. During meiotic pairing the chromosome bearing the duplicated segment forms a loop. Pairing and exchange (crossing over) in inverted and displaced duplications leads to different secondary chromosome structural variants (i.e., chromosomal aberrations) such as reciprocal translocation, inversion, rings, acentric and dicentric chromatids. 128 Duplication and deletion (after Sutton, 1965).



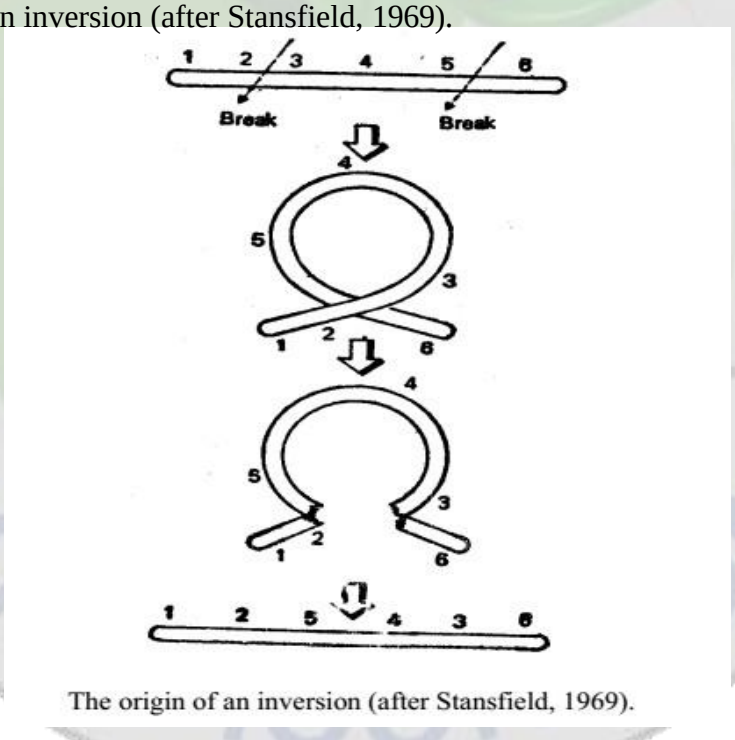
Duplication and deletion (after Sutton, 1965).

Genetic significance of Duplications 1. The duplications of chromosomes are not deleterious to the organism like the deficiency, but, they usually protect the organism from the effect of a deleterious recessive gene or from an otherwise lethal deletion. 2. some

duplications are useful in the evolution of new genetic material. In an organism with duplications, because the old genes can continue to provide for the present requirements of the organism, the superfluous genes may be free to mutate to new forms without a loss in immediate adaptability. 3. Large duplications can reduce the fertility as a result of meiotic complication, and in this way reduce their own probability of survival (Sybenga, 1972). 4. Relocation of chromosomal material without altering its quantity may result in an altered phenotype, this is called position effect. 129

3. Inversion:

An inversion is an intra-chromosomal aberration in which a segment is inverted 180 degrees. For example if a chromosome has segments in the order of 1-2-3-4-5-6 and breaks occur in regions 2-3 and 5-6 and the broken piece (3-4-5-) is reinserted in reverse order, then the inverted chromosome will have segments in order of 1-2-5-4-3-6, such as shown in the figure 21.5: The origin of an inversion (after Stansfield, 1969).



The origin of an inversion (after Stansfield, 1969).

In a diploid organism, when out of two homologous chromosomes one chromosome undergoes the inversion, then, it is called inversion heterozygote. During synapsis of such a homologous pair having inversion heterozygoe, the synapsis configuration attempts to maximize the pairing between homologous regions in the two chromosomes. This is usually accomplished by a characteristic inversion loop in one of the chromosome. 130

Types of inversions

The inversions are of following types:

i) Pericentric inversions – When the inverted segment of chromosome includes or contains centromere, then such inversions are called heterobranchial or pericentric inversions. If crossing over occurs with in the loop of a pericentric inversion, the resulted chromatids include half on the chromatids with duplications and deficiencies forming nonfunction. The other half of the chromatids form functional gametes: ¼ gametes have normal chromosome order, ¼ gametes have the inverted arrangement.

ii) Paracentric inversions – When the inverted segment includes no centromere and the centromere remains located outside the segment, then such type of inversion is called homobranchial or paracentric inversion. Crossing over within the inverted segment of a paracentric inversion, produces a dicentric chromosome contains two centromeres and forms a bridge from one pole to the other during first meiotic anaphase. When anaphase chromosomes separate towards poles, this bridge breaks somewhere along its length and the resulting fragments contain duplications and/ or deficiencies. The acentric chromosome because lacks in centromere and fails to move to either pole and so, is not included in the meiotic products. Such, breakage-fusion bridge cycles of crossing over of paracentric inversions are most common in maize. The meiotic products includes half non-functional, ¼ functional normal and ¼ functional inverted chromosomes. Crossing over in paracentric and pericentric inversions (after SRB, Owen and Edger, 1965) 131

Genetic significance of inversions

(i) Simple inversions do not have primary phenotypic effects other then on chromosome shape. Frequently, however, some DNA at a break point has been damaged and this may result in an observable mutation, often recessive (e.g., c 1B lethal mutation in Drosophila).

(ii) Due to inversion a peculiar kind of position effect occurs. The position effect is caused by the transfer of a gene from a euchromatic segment to the vicinity of heterochromatic segment. Heterochromatinization may then extend into a displaced, originally euchromatic region and suppress the transcription of the gene in it.

(iii) Normal linear pairing is not possible in inversion heterozygotes. The difficulties encountered with pairing cause a reduction of exchange (crossing over) in and around the inversion.

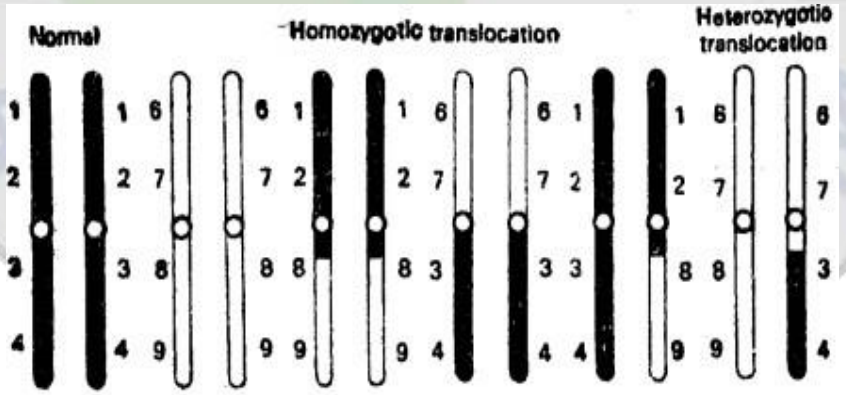
(iv) They maintain heterozygosity from generations to generations.

B. Interchromosomal aberrations When breaks occur in non-homologous chromosomes and resulting fragments are interchanged by both of the non-homologous chromosomes, the inter-chromosomal or heterosomal aberrations occur. The inter-chromosomal aberration is of following type:

Translocation

Translocation involves the shifting of a part of one chromosome to another nonhomologous chromosome. If two non-homologous chromosomes exchange parts, which need not be of the same size, the result is a reciprocal translocation. The reciprocal translocation may be of following types:

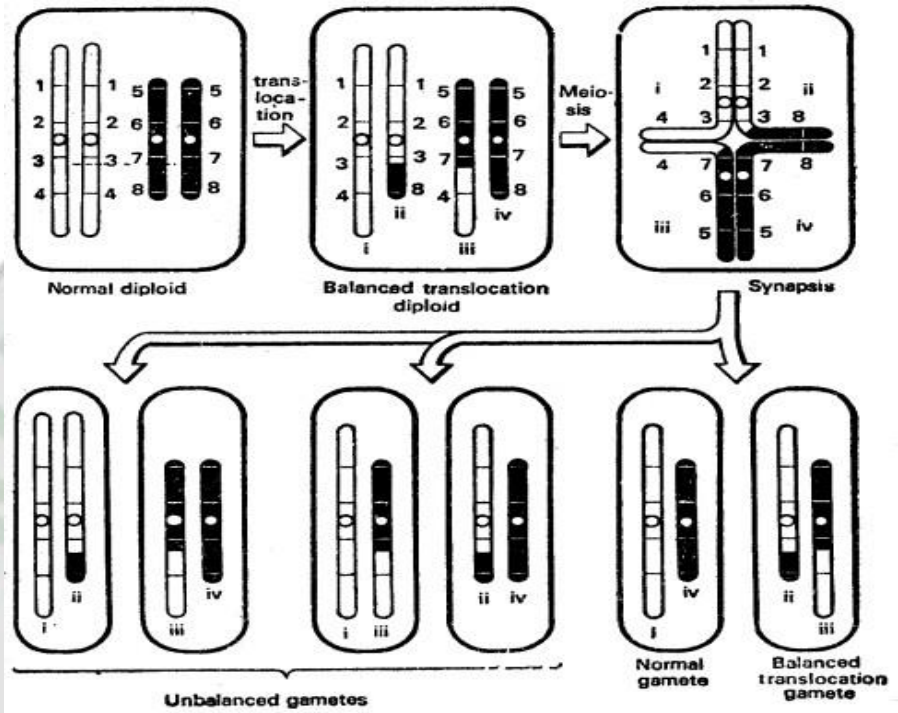
1. Homozygotic translocation - In homozygotic translocation normal meiosis occur and cannot be detected cytologically. Genetically they are marked by altered linkage group by the fact that a gene with new neighbours may produce a somewhat different effect in its new location (position effect). Homozygotic and heterozygotic translocations (after De Robertis, Saez and Nowinski 1970).



Homozygotic and heterozygotic translocations
(after De Robertis, Saez and Nowinski 1970)

2. Heterozygotic translocation – In heterozygotic translocation a considerable degree of meiotic irregularity occur. During meiosis, an individual which is heterozygous for a reciprocal translocation must form a cross-shaped configuration in order to affect pairing of

all homologous segments. This cross-shaped configuration often opens out into a ring as chiasmata terminalize. The meiotic products (gametes) are of three types –normal balanced and unbalanced gametes as have been illustrated in following diagram: Meiosis in heterozygotic translocation (after Sutton 1965).



Meiosis in heterozygotic translocation (after Sutton 1965).

Genetic significance of Heterozygotic Translocation:

1. The heterozygous translocation produce semi-sterile organisms because between half and two third gametes fail to receive the full complements of genes required for normal development of sex.

2. Some genes which formerly assorted independently, exhibit linkage relationships after translocation has occurred; a single reciprocal translocation will reduce the number of linkage groups by one.

3. The phenotypic expression of a gene may be modified when it is translocated to a new position in the genome (position effect).

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