Chapter 13 THERMOREGULATION AND OSMOREGULATION

STUDENTS'LEARNING OUTCOMES

After studying this chapter, the students will be able to:

- Describe three elements i.e., receptors, control center and effectors which operate homeostatic mechanisms.
- Relate the homeostatic mechanisms with the negative and positive feedback systems.
- Define thermoregulation and explain its needs.
- Classify animals on the basis of the source of body heat i.e., ectotherms and endotherms.
- Classify the animals on the basis of the ability to thermoregulate i.e., poikilotherms and homeotherms.
- Describe the regulatory strategies in man for thermoregulation.
- Differentiate between osmoconformers and osmoregulators.
- Define osmoregulation.
- Explain the problems faced by osmoregulators.
- Explain the different methods of osmoregulation found in freshwater, marine water and terrestrial animals.

In this chapter, we will study homeostasis. We will explore the mechanisms of thermoregulation and osmoregulation and examine the importance of maintaining homeostasis for the health and survival of human beings.

13.1- Homeostasis

Homeostasis is a vital process by which living organisms maintain a stable internal environment, despite changes in their external environment. Homeostasis consists of a wide range of mechanisms that work together to regulate the body's internal environment. Important processes of homeostasis are thermoregulation, osmoregulation, and excretion.

13.1.1- Basic Mechanism of Homeostasis

Homeostasis is maintained through the collaboration of three essential elements i.e., receptors, control centre, and effectors.

1- Receptors

Receptors or sensors are specialized structures (organs, tissues, cells or subcellular components) that detect changes in the internal or external environment. They also transmit the information of changes to the control centre. For example,

receptors cells present in the skin can detect changes in temperature. Similarly, receptors cells in the blood vessels can detect changes in blood pressure and pass information of the control centre.

2- Control Centre

The control centres receive information from the receptors, process this information, and send appropriate message to the effectors. These messages ensure that the internal environment remains within a narrow range of set points. These set points are optimal for the body's function. The brain, spinal cord and endocrine glands act as control centres in homeostasis.

3- Effectors

Effectors are the parts of the body which respond when they receive message from the control centres. Muscles and glands act as effectors. Muscles respond by contracting while glands release their secretion. In this way, the responses of effectors restore the internal environment to its set points. For example, when the body temperature drops below the set point, effectors in the skin and muscles cause shivering and vasoconstriction. This response increases heat production and raises the body temperature to the set point.

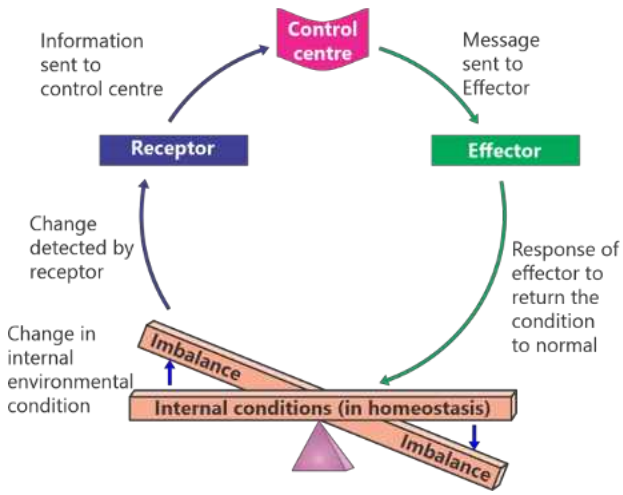


Figure 13.1: Elements of homeostasis

13.1.2- Feedback Mechanisms

Feedback mechanisms are the foundations of homeostasis. These are the regulatory processes in which the output or product of a system is used to control the behavior of the system itself. There are two types of feedback mechanisms.

Negative Feedback

A negative feedback system is a mechanism in which the output or product of a system reduces the activity of the system. It is the most common form of regulation. Common examples of negative feedback include the regulation of body temperature, blood glucose levels, and hormone secretion.

For example, if body temperature exceeds the set point of 37°C, the thermoreceptors in the hypothalamus of brain detect this change. They send signals to the control centre (thermoregulator in hypothalamus). The control centre sends messages to effectors (e.g., sweat glands). The effectors respond by producing sweat which lowers the temperature. The output of this process (lowering of temperature to the set point) influences the thermoreceptors negatively. As a result, the thermoreceptors stop sending message to the control centre.

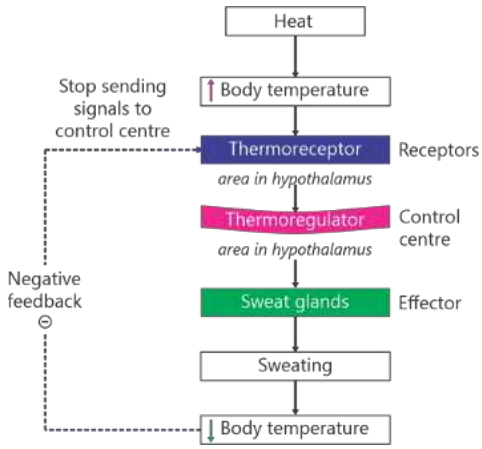


Figure 13.2: Negative feedback system to control body temperature

Positive Feedback

A positive feedback system is a regulatory mechanism in which the output or product feed increases the activity of the system. In this way, the positive feedback enhances the original stimulus. In homeostasis, positive feedback is less common than negative feedback. It often happens in the processes that require rapid and significant changes e.g., blood clotting, labour and childbirth.

For example, during childbirth, contractions of the uterus send nerve impulses to the control centre (part of hypothalamus). The control centre sends signals to pituitary gland to release oxytocin hormone. Oxytocin then stimulates further contractions of the uterus, which in turn sends more nerve impulses to hypothalamus, leading to the release of more oxytocin. This positive feedback loop continues until the baby is born.

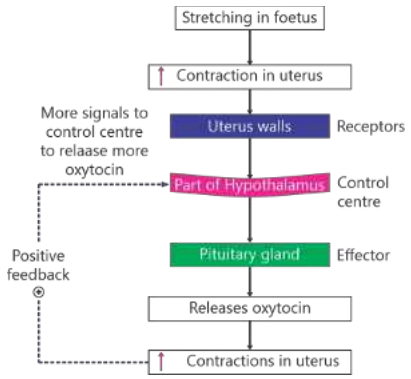


Figure 13.3: Positive feedback system to in childbirth

Unregulated Positive Feedback

Sometimes, the un-regulated positive feedback systems can result in health problems. For example; in severe inflammation or infection, the immune cells release proteins called cytokines. These proteins further stimulate the immune response. If this positive-feedback is not properly regulated, it can lead to excessive inflammation and tissue damage.

13.2- Osmoregulation

The maintenance of the balance of water and solutes in the body fluids is called osmoregulation. It ensures that the body fluids do not become too dilute or too concentrated. It is essential for many physiological processes, including maintaining proper blood pressure, enabling the proper function of organs, and regulating metabolism.

Recalling

- Osmosis is the diffusion of water across a membrane, and it always occurs from a more dilute (hypotonic) solution to a less dilute (hypertonic) solution.
- The osmotic pressure of a solution is a measure of its tendency to take in water by osmosis.

13.2.1- Osmoregulators and Osmoconformers

On the basis of the ability of osmoregulation, the animals are divided into two groups i.e., osmoconformers and osmoregulators.

Osmoconformers

Osmoconformers are the animals which allow the osmotic concentrations of their body fluids to match their environment. They cannot maintain the concentration of water and solutes in their bodies. So, these concentrations change with the changes in surrounding medium and they remain isotonic to their environment. Such animals are usually found in environments with relatively stable solute concentrations, such as marine water. Examples of osmoconformers include all marine invertebrates, some freshwater invertebrates and some marine vertebrates like hag fishes, sharks and rays.

Osmoregulators

The animals that can regulate the concentrations of water and solute in body fluids are osmoregulators. They can maintain constant internal concentrations despite the changing concentration in the environment. They use various mechanisms for osmoregulation e.g., active transport of salts, accumulation of salts, and water conservation etc. Most vertebrates in aquatic and terrestrial environments are osmoregulators.

13.2.2- Problems and Methods of Osmoregulation

Osmoregulation in Freshwater

Generally, the freshwater animals are hypertonic to their environment. It means that they have higher solute concentration in body fluids than the surrounding water. Water continuously enters their bodies through osmosis, particularly across permeable surfaces like gills. Essential salts and ions (like Na^+ and Cl^-) diffuse out of the body into the surrounding water.

In osmoregulation, they prevent water from entering their bodies and eliminate the excess water that does enter. For this purpose, they produce large volumes of diluted urine. To compensate the loss of salts, they actively transport salts back into their bodies. Unlike their marine water animals, freshwater animals rarely drink water.

Osmoregulation in Marine Water

Most marine animals are hypotonic to their environment. This means that external environment has a much higher solute (salt) concentration than their internal tissues. Water constantly leaves their bodies via osmosis through the gills and skin. Salts continuously diffuse from the environment into their bodies. In addition, they have excess of salts in the body due to drinking of sea water.

To compensate for the water loss, marine animals drink seawater continuously. They also conserve water by producing very small amounts of highly concentrated urine. To remove the excess salts, specialized chloride cells in the gills use energy (ATP) to actively pump salts out of the blood and back into the environment. They also remove excess salts through their kidneys. Some marine fishes also have special salt excreting glands in rectum called rectal glands. These glands remove salts from blood into the digestive tract which are then eliminated along with faeces.

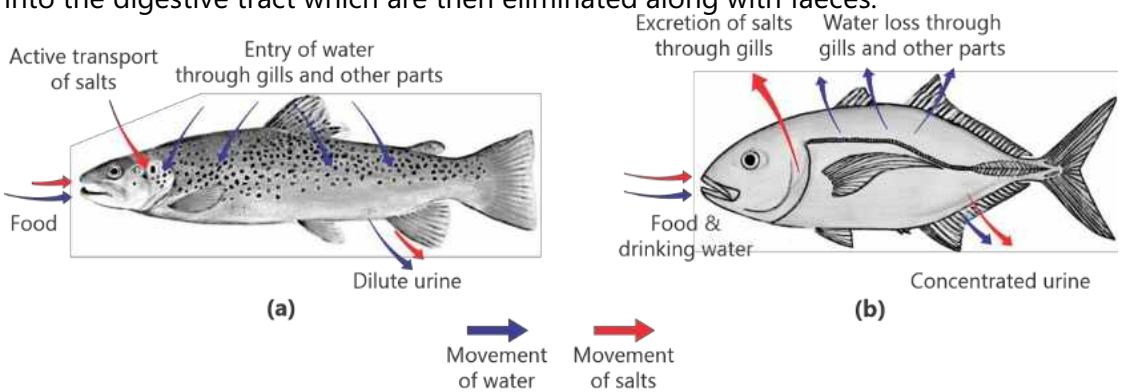


Figure 13.4: Osmoregulation in (a)- freshwater and (b)- marine water

Osmoregulation in Terrestrial Habitats

Terrestrial animals lose water to the environment through evaporation from respiratory surfaces, through the skin, and via the excretion of waste. They have to ensure that water intake (from drinking and food) and metabolic water production is equal to the water lost to the environment. These animals have anatomical, physiological, and behavioral adaptations to maintain osmotic balance.

They have relatively impermeable outer layers to prevent evaporative water loss. For example, insects possess a waxy exoskeleton made of chitin. Reptiles have waterproof scales made of keratin. Mammals and birds have dead, keratinized skin cells and also have oil secreting glands in skin.

Their kidneys play a vital role to retain water inside the body. Moreover, they convert the primary waste product of protein metabolism i.e., ammonia into urea or uric acid which require less amount of water for excretion.

Many desert animals stay underground during the heat of the day to avoid evaporation. They emerge only at night when the air is cooler and more humid. These animals feed upon seeds of desert plants in which large amount of carbohydrate are stored. Some animals, like the Kangaroo Rat, survive entirely on metabolic water (produced as a by-product of cellular respiration).

Anhydrobiosis:
Certain invertebrates, like tardigrades (water bears), enter a dormant state when they lose almost all their body water. They come to active life years later when water returns.

13.2.3- Nature of Excretory Products in relation to Habitat

In many animals, osmoregulation is coupled with the removal of metabolic wastes from the body. The nitrogenous wastes (e.g., ammonia, urea, uric acid) are the main metabolic wastes. These wastes are excreted as water solutes. The animals of different types of habitats excrete different forms of nitrogenous wastes. For example;

1- Ammonia – NH_3

It is produced when amino group ($-NH_2$) is removed from amino acids and nucleic acids and is combined with H^+ to form ammonia (NH_3) in the liver. Ammonia is highly toxic and so it is excreted rapidly. One gram of ammonia requires 500 mL of water to dissolve it to nontoxic level. Such plenty of water can only be afforded by freshwater animals e.g., most fishes, protozoans, sponges, coelenterates. The marine animals excrete ammonia directly into the water. Animals which excrete ammonia as their major nitrogenous wastes are called **ammonotelic**.

2- Urea - $CO(NH_2)_2$

As the terrestrial animals face the challenge of conserving water, they cannot afford to excrete ammonia with a plenty of water. They convert ammonia to urea in their liver and then excrete urea through kidneys. Urea is less toxic than ammonia. One gram of urea requires 50 mL water to dilute it to the nontoxic level. The animals which excrete urea as their major nitrogenous waste product are called **ureotelic**.

Humans also excrete a small amount of uric acid in urine. Uric acid is a by-product of the breakdown of purines (found in many foods, particularly meat and some types of vegetables).
Uric acid is produced in the liver. In healthy individuals, most of the uric acid produced is excreted in the urine, but when there is an excess of uric acid in the blood, it can lead to the formation of uric acid crystals in joints and tissues, causing painful conditions such as gout.

3- Uric acid - $C_5H_4N_4O_3$

Some animals, such as insects, desert animals, birds and most reptiles face an extreme water conservation challenge due to the arid nature of their environment. They cannot afford to even excrete urea. They convert ammonia to uric acid by using lot of energy. Uric acid is even less toxic than urea. It is less soluble in water and makes precipitates. One gram of uric acid requires just 1 mL of water for its excretion. The animals which excrete uric acid are called **uricotelics**.

13.3- Thermoregulation

Thermoregulation is the homeostatic process by which organisms maintain their internal body temperature within a tolerable range, regardless of the temperature of the external environment.

Thermoregulation is very important for animals. We know that enzymes have an optimal temperature at which they function most efficiently. If temperature falls below the optimal, enzyme-substrate collisions decrease, and metabolic rates drop. If temperatures rises well above the optimal, enzymes begin to denature and lose their function permanently. Similarly, the lipid bilayer of cell membranes is temperature-sensitive. At high temperatures, cell membranes can lose their selective permeability. Low temperatures can make membranes rigid and brittle.

13.3.1- Types of Animals on the basis of Heat Source

Ectotherms

Ectotherms are animals that gain most of their body heat from external sources. Their internal temperature generally fluctuates with the temperature of their surroundings. Most invertebrates are ectotherms. Among the vertebrates, fishes, amphibians, and reptiles are ectotherms. In ectotherms, the metabolism rate is very low. They cannot produce enough heat to raise and maintain their body temperature. So, they rely on the heat from the environment. Ectotherms use behavioral thermoregulation. If they are too cold, they bask in the sun (radiation) or sit on a warm rock (conduction). If they are too hot, they seek shade or burrow into the cool earth. In very cold climates, they become dormant during the coldest months.

Endotherms

Endotherms generate the majority of their body heat through internal metabolism. They maintain a relatively constant body temperature, often higher than the environment. Birds and mammals are endotherms. When it gets cold, they may shiver (muscle contractions) or burn brown adipose tissue for heat. Most endotherms have insulation to retain heat such as hair, feathers, or fat.

13.3.2- Types of Animals on the basis ability of Thermoregulation

Poikilotherms

Poikilotherms are the animals that are unable to maintain their body temperature within narrow limits. Their body temperature varies considerably, usually matching the temperature of environment. All invertebrates are poikilotherms. The vertebrates: fishes, amphibian and reptiles are also poikilotherms.

Poikilotherms do not need to consume massive amounts of food to "fuel" a constant temperature. It allows them to survive in nutrient-poor environments where a homeotherm would starve.

Homeotherms

These animals maintain relatively constant internal body temperatures, regardless of environmental fluctuations. They use "thermostats" (hypothalamus in the brain) to trigger cooling or heating responses. Examples include mammals and birds. For instance, humans maintain their internal temperature at roughly 37°C whether they are in very cold or very hot environments.

13.3.3- Thermoregulation in Humans

Humans are endothermic homeotherms. They regulate body temperature through negative feedback mechanism. For example; the thermoreceptors present in skin detect an increase or decrease in body temperature. They send signals to the control centre i.e., hypothalamus which also contains thermoreceptors. Hypothalamus sends signals to muscles and glands which produce corrective responses e.g.,

Responses to Overheating (Hyperthermia)

When body temperature rises above 37°C (hyperthermia), vasodilation occurs. It is the expansion of blood capillaries which lie just beneath the epidermis of the skin. So, there is more flow of blood in these capillaries to remove more heat from blood. Responses also include sweating. Evaporation of sweat carries heat from the body and produces cooling effect. The thyroid gland may decrease the secretion of thyroxine, which lowers the basal metabolic rate. So, the internal heat production is reduced.

The thermoreceptors also stimulate the higher centres of brain. So, we 'feel' warmth or coldness. It allows us to take more actions like taking a bath or turning the fans or heaters on.

Responses to Cooling (Hypothermia)

When body temperature falls below 37°C (hypothermia), the responses may include vasoconstriction, shivering and increase in metabolic rate. Vasoconstriction reduces blood flow to the skin and so there is less loss of heat from blood. Shivering (spasmodic contraction of skeletal muscles) produces heat which helps to raise the

body temperature. Tiny muscles at the base of hair follicles contract (goosebumps). Air is trapped in the erected hair and act as insulator for the heat. Increased metabolic rate also produces heat to raise the body temperature.

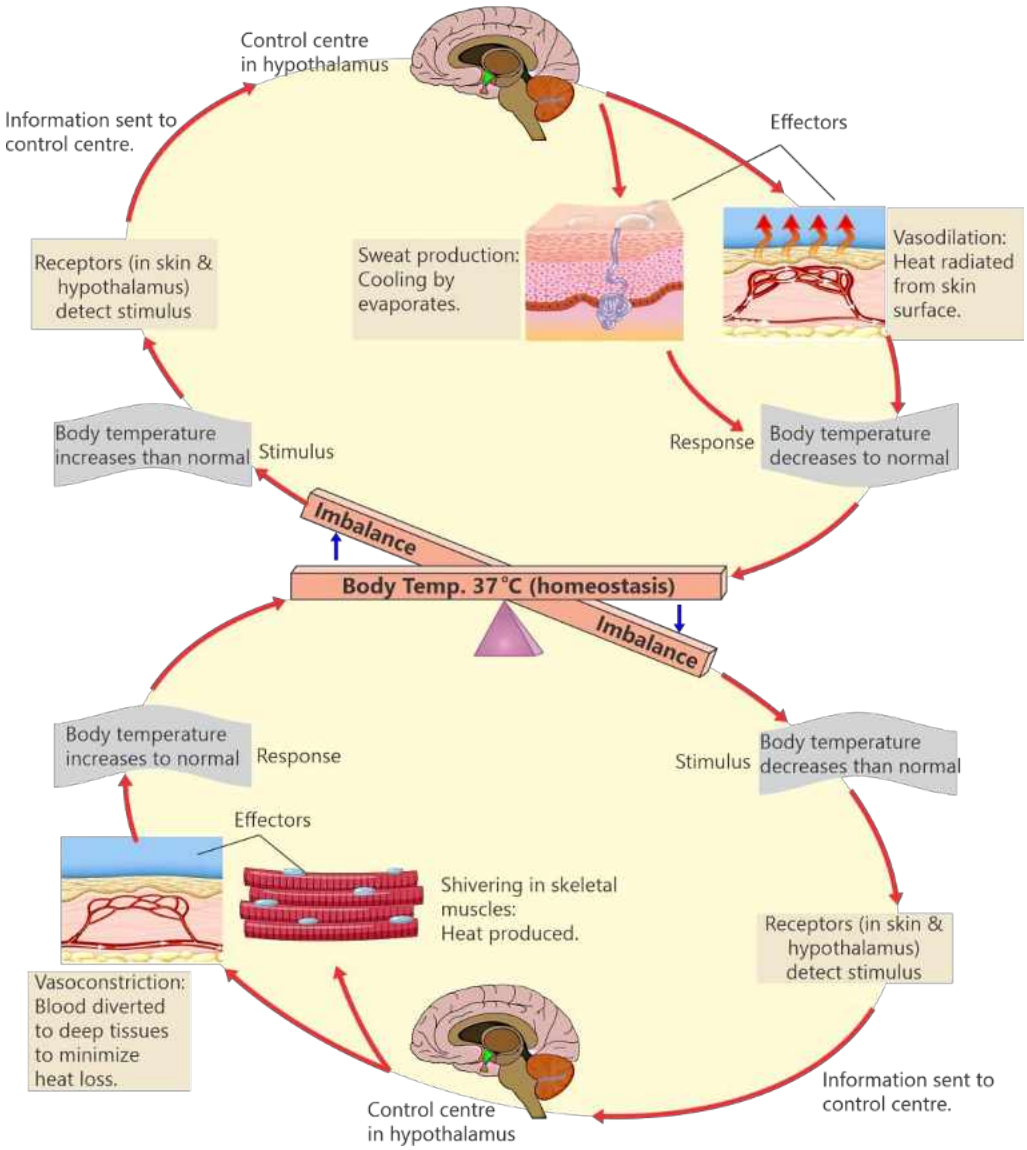


Figure 13.5: Thermoregulation in humans

EXERCISE

SECTION 1: MULTIPLE CHOICE QUESTIONS

1. Which term covers the rest of the three?
   (a) Osmoregulation (b) Excretion
   (c) Thermoregulation (d) Homeostasis
2. Marine fishes compensate for water loss by:
   (a) Producing dilute urine (b) Drinking seawater
   (c) Absorbing salt (d) Reducing thirst
3. How do freshwater fish primarily gain water?
   (a) Drinking (b) Active transport
   (c) Osmosis (d) Diffusion
4. What is the primary heat source for endotherms?
   (a) Sunlight (b) Internal metabolism
   (c) Warm rocks (d) Convection
5. What is the control center for human thermoregulation?
   (a) Pituitary gland (b) Hypothalamus
   (c) Adrenal gland (d) Medulla oblongata
6. Which waste product requires the most water for excretion?
   (a) Ammonia (b) Urea
   (c) Uric acid (d) Creatinine
7. Which nitrogenous waste is the least toxic and water-insoluble?
   (a) Urea (b) Ammonia
   (c) Uric acid (d) Nitrate
8. In which part of the body, the conversion of ammonia to urea takes place?
   (a) Liver (b) Muscles
   (c) Brain (d) Small intestine

SECTION 2: SHORT QUESTIONS

1. Define osmoregulators and osmoconformers.
2. Define thermoregulation and explain its needs.
3. What are the two types of animals on the basis of the ability to thermoregulate?
4. Differentiate between:
   - Osmoregulators and osmoconformers
   - Ectotherms and endotherms
   - Vasodilatation and vasoconstriction

- Homeotherms and poikilotherms
- Ammoniotelic and ureotelic
- Ureotelic and uricotelic

SECTION 3: LONG QUESTIONS

1. Describe three elements which operate homeostatic mechanisms.
2. Relate the homeostatic mechanisms with the negative and positive feedback systems.
3. Explain the different methods of osmoregulation found in freshwater, marine water and terrestrial habitats.
4. Explain the nature of nitrogenous wastes in relation to habitat.
5. Classify animals on the basis of the source of body's heat.
6. Classify the animals on the bases of the ability to thermoregulate.
7. Describe the regulatory strategies in man for thermoregulation.

INQUISITIVE QUESTIONS

1. Why positive feedback mechanisms in humans are sometimes associated with severe health problems?
2. List some of the behavioural responses of the animals to maintain homeostasis.
3. Draw a flow chart to show negative feedback of homeostatic mechanisms by taking an example of hormone.

Chapter 14 HUMAN URINARY SYSTEM

STUDENTS'LEARNING OUTCOMES

After studying this chapter, the students will be able to:

- Outline different organs of the (human) urinary system.
- Describe the structure of the kidney.
- Relate the structure of the kidney with its function.
- Explain the detailed structure of a nephron.
- Explain the processes of glomerular filtration, selective re-absorption and tubular secretion as the events in kidney functioning.
- Explain regulatory mechanism involved in concentration of urine
- Justify the functioning of kidneys as both excretion and osmoregulation.
- Compare the function of two major capillary beds in kidneys i.e., glomerular capillaries and peritubular capillaries.
- List urinary tract infections and the bacteria responsible.
- Explain the causes and treatments of kidney stones.
- Outline the causes of kidney failure.
- Explain in detail the mechanism and problems related to dialysis.
- Describe the principles and the problems associated with kidney transplant.

In this chapter, we will explore the remarkable architecture and functioning of the kidneys. We will also study the life-saving technologies used when this vital system weakens.

14.1- Human Urinary System

The urinary or excretory system is responsible for excretion. In biological terms, **excretion** is the process by which an organism eliminates the metabolic wastes. The metabolic wastes include nitrogenous wastes, extra water, and extra salts etc. The urinary system performs three essential roles simultaneously:

Critical Thinking
Given the definition of *excretion*, why the large intestine is not classified as a major excretory organ?

1. **Nitrogenous Waste Removal:** Filtering out urea, a nitrogenous waste of protein breakdown in the liver.
2. **Osmoregulation:** Maintaining the "water-to-salt" balance of the blood by adjusting how much water is reabsorbed or excreted.
3. **pH and Ion Regulation:** Balancing levels of sodium, potassium, and hydrogen ions to keep blood slightly basic (around pH 7.4).

The human urinary system consists of two kidneys, two ureters, one urinary bladder, and one urethra. Kidneys are located high in the abdominal cavity, one on each side of the vertebral column, between the 12th thoracic and 3rd lumbar vertebrae. Due to the position of liver, the right kidney is slightly lower and smaller than the left one. The upper parts of both kidneys are partially protected by the 11th and 12th ribs. The **ureters** are about 25 cm long tubes. A ureter begins at the funnel-shaped renal pelvis in kidney. Ureters from both kidneys extend downward and open in a single urinary bladder. Ureters transport urine from the kidney to the **urinary bladder**. The urinary bladder is a hollow and distensible muscular organ. It is located within the pelvic cavity. It serves as urine reservoir. The **urethra** is a tube that carries urine from urinary bladder to the outside of the body.

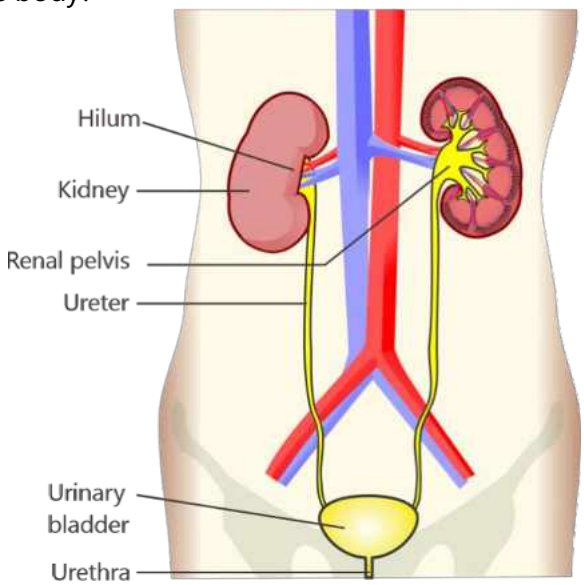


Figure 14.1: Human excretory (urinary) system

14.1.1- Structure of Kidney

A kidney is a reddish brown, bean shaped organ. Each kidney, has an adrenal gland on its top. The kidney and its adrenal gland are surrounded by a fibrous **renal capsule**. Each kidney has a convex and a concave side. A depressed area on the concave side is the **renal hilum** (also called hilus). At this point, the renal artery enters the kidney and the renal vein and ureter leave.

Internally, a kidney consists of two portions. The outer portion is called **renal cortex**. It makes up about a third of the kidney's mass. The inner portion is called **renal medulla**. It makes two-thirds of the kidney. The **renal pelvis** is a funnel-shaped structure in the centre of kidney. Blood enters the kidney through a renal artery and leaves through a renal vein. The renal medulla consists of a number of cone-shaped **renal pyramids**. Urine is collected in the renal pelvis and exit the kidney through ureter.

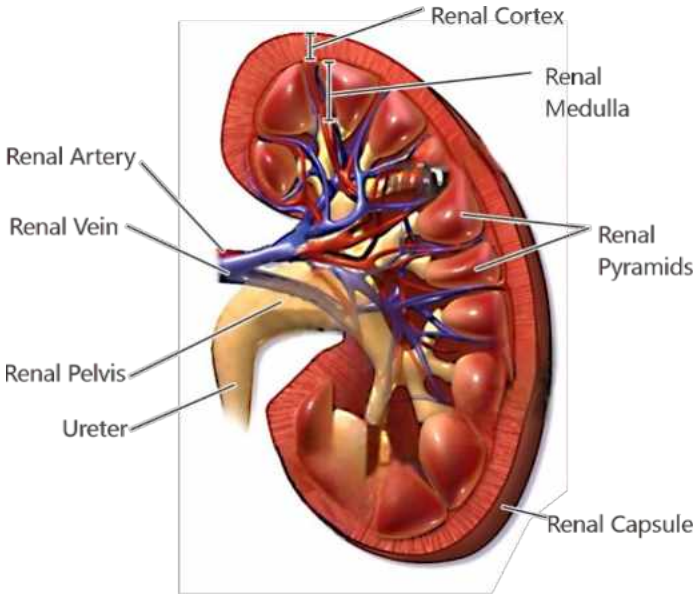


Figure 14.2: The internal structure of kidney

Structure of Nephron

Each kidney contains about one million microscopic tubular structures called nephrons. These are the functional units of kidney. A nephron consists of two main portions i.e., a renal corpuscle and a renal tubule.

Renal corpuscle consists of two associated structures. **(i) Glomerulus** is a capillary network derived from an afferent renal arteriole. **(ii) Bowman's capsule** is a cup-shaped, double-walled sac that surrounds the glomerulus.

Renal tubule is a long tubule that leads away from the Bowman's capsule. It consists of three connected portions. **(i) The proximal convoluted tubule** is the beginning portion and is highly coiled. It dips into the medulla. **(ii) The Loop of Henle** is a U-shaped hairpin turn. It consists of two limbs. The descending limb continues toward the medulla. The ascending limb returns toward the cortex and reaches parallel to renal corpuscle. Here, it opens in **(iii) the distal convoluted tubule** which is highly coiled again.

The distal convoluted tubules of multiple nephrons drain into a single **collecting duct**. As these ducts descend through the medulla, they merge to form larger **papillary ducts**, which ultimately discharge into the renal pelvis.

Nephrons are categorized into two types based on their location within the kidney: **(i) Cortical nephrons** make the majority (about 85%) of nephrons. These are located primarily within the renal cortex. They possess relatively short loops of Henle. **(ii) Juxtamedullary nephrons** originate deep in the cortex, near the medulla. They have long loops of Henle that extend deep into the renal medulla.

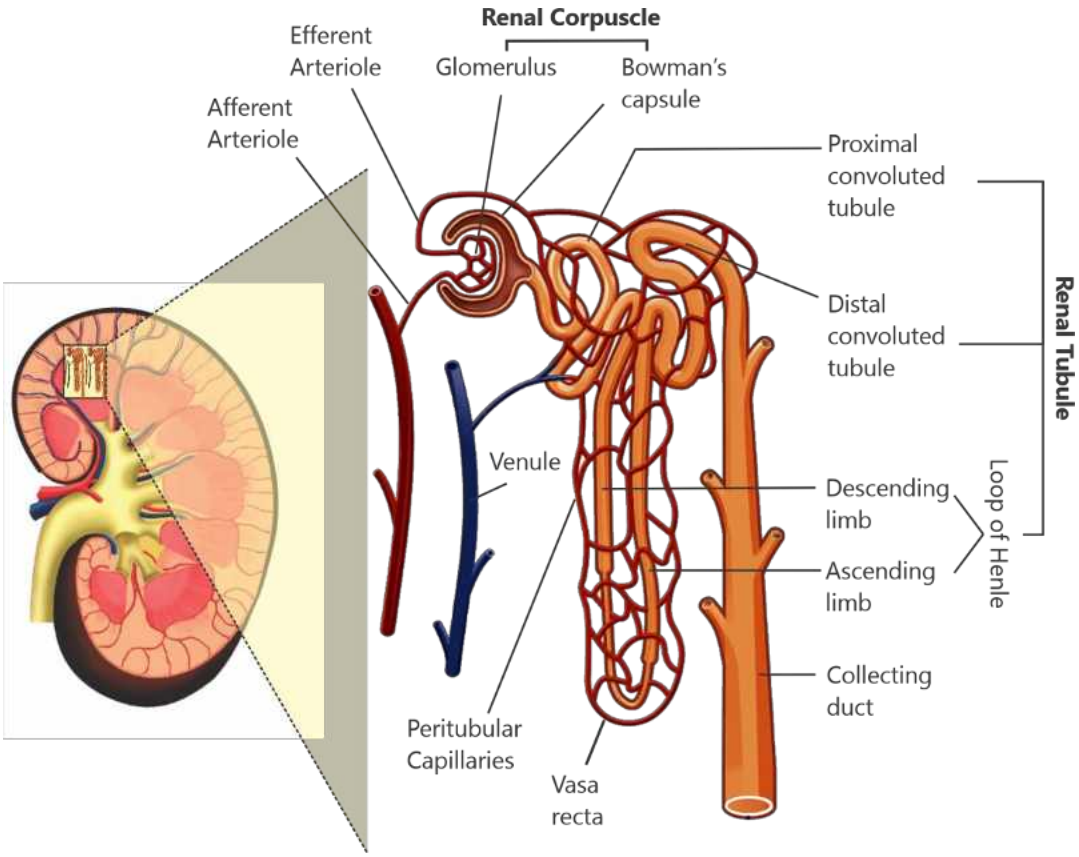


Figure 14.3: Structure of nephron

14.1.2- Blood Supply of the Kidney

The pathway of blood inside kidney begins with the **renal artery**, which enters the kidney at the hilum. It divides into branches which give rise to **afferent arterioles**. Each afferent arteriole leads into the capillary bed i.e., glomerulus. Unlike most capillary beds in the body that drain into venules, the glomerular capillaries drain into an efferent arteriole.

> The kidneys are highly vascularized organs, receiving approximately 20–25% of the total cardiac output.

The **efferent arteriole** branches into **peritubular capillaries**. These capillaries form a dense network around the proximal and distal convoluted tubules in the renal cortex. In juxtamedullary nephrons, the efferent arterioles also give rise to **vasa recta**. These are long and straight capillary networks in medulla that run parallel to the loops of Henle.

After the exchange of materials, the capillaries converge to form **venules**. These venules merge into larger veins, eventually opening into the **renal vein**. The renal vein exits the kidney at the hilum and drains into the inferior vena cava. Here it returns the filtered, "clean" blood to the heart.

14.1.3- Functioning of Kidney

Kidneys filter metabolic wastes and extra water from blood and excrete them in the form of urine. The process of urine formation comprises three major steps that take place in the nephron: glomerular filtration, selective reabsorption, and tubular secretion.

While doing the excretory function, kidneys also work as osmoregulatory organs. Kidneys control the salt concentrations of body fluids by forming dilute urine (when body fluids are hypotonic) and concentrated urine (when body fluids are hypertonic and there is dehydration).

(i)- Glomerular Filtration

The first step in the formation of urine is a physical process known as glomerular filtration (or **ultrafiltration**). The efferent arteriole is narrower than the afferent arteriole. Due to this difference, the blood within the glomerular capillaries remains under relatively high pressure. This high-pressure in glomerulus forces water, urea, glucose, amino acids, vitamins, and various salts ((Na^+, K^+, Cl^-) from blood into the Bowman's capsule. About one-fifth (20%) of the fluid portion of the blood filters into the Bowman's capsule and is called **glomerular filtrate**. Plasma proteins (e.g., albumin and globulins) and blood cells are too large to pass through the capillary walls. So, these structures remain in the blood.

(ii)- Selective Reabsorption

The glomerular filtrate contains not only wastes but also vital nutrients that the body cannot afford to lose. Selective reabsorption is the process by which useful materials are transported from renal tubule back into the peritubular capillaries. Most reabsorption occurs in the proximal convoluted tubule. In this region, about 75% of the water returns from the filtrate to the capillaries by osmosis. Glucose, amino acids, and salts (sodium, potassium, and calcium) are returned from proximal tubule to the blood by active transport. As the filtrate moves further, more water is reabsorbed in the descending limb of the loop of Henle. Ascending limb is impermeable to water but actively transports salts out of the tubule. Some water and salts are also absorbed from distal convoluted tubule. Collecting duct is the final site for water reabsorption.

(iii)- Tubular Secretion

Tubular secretion is opposite to reabsorption. In this process, the epithelial cells of the distal convoluted tubule take some materials from surrounding blood and secrete them into the filtrate. Secreted substances include hydrogen ions, ammonium, potassium ions, and various metabolic toxins or drug residues (like penicillin). Through tubular secretion of hydrogen ions, the kidneys play role in maintaining the acid-base balance (pH) of the blood.

Elimination of Urine

From the collecting ducts, urine flows through the renal pelvis into a ureter. Ureter leads to the urinary bladder. Muscular contractions of the bladder force urine out of the body

through urethra. A normal adult eliminates from 1 to 2 L of urine a day. This amount is highly variable and depends on several factors. The more water or hydrating fluids we consume, the higher is the urine output. Diets with high salt trigger water retention and decrease urine production. Physical exercise leads to water loss through respiration and perspiration, which generally decreases urine volume.

Tidbit

In hot weather, the body loses more water through sweat, leading the kidneys to conserve water and produce a smaller volume of concentrated urine.

14.2- Regulation of the Urine Concentration

14.2.1- Counter-current Mechanism

For the reabsorption of water from the loop of Henle and collecting duct, nephrons create and maintain higher concentration of salts (NaCl) in the surrounding fluid. It is done by **counter-current mechanism**.

- The descending limb is permeable to water but not to salts.
- The ascending limb is impermeable to water but actively transports salts out of tubule.

When filtrate flows through the descending limb, water is reabsorbed into the surrounding fluid because the surrounding fluid has more salts. When fluid flows through the ascending limb, it actively transports sodium ions from the filtrate to the surrounding fluid. The chloride ions follow sodium ions. So, the salt concentration of the fluid remains high As urine passes down the collecting duct through this salty fluid, water moves out by osmosis and enters blood via the vasa recta.

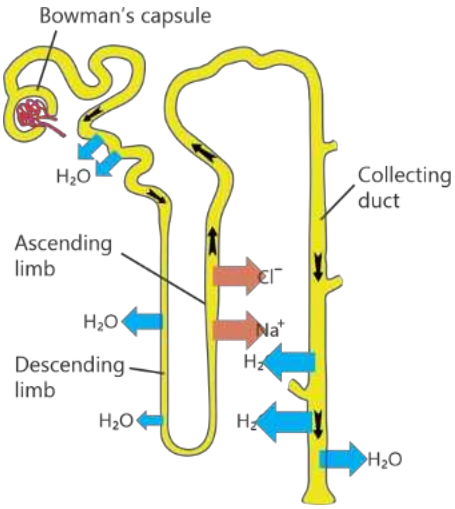


Figure 14.4: Formation of urine

14.1.2- Hormonal Control of Kidney Functions

When body needs to conserve water, kidneys excrete a hypertonic urine.When there is much water in body, kidneys excrete a hypotonic urine. These functions of kidneys are coordinated by the following hormones.

1- Antidiuretic Hormone (ADH)

ADH is the body's primary tool for water conservation. It is produced by the **hypothalamus** and secreted by the **posterior pituitary gland**

When the body is dehydrated or has taken salty food, the higher concentration of salts in blood is detected by osmoreceptors present in the hypothalamus. The

hypothalamus sends signals to the posterior pituitary to create a sensation of thirst and to release ADH. ADH acts on the walls of the collecting ducts and make them more permeable to water. So, more water moves from the filtrate into the hypertonic fluid and then into blood. In this way, concentrated urine is produced and water is conserved.

When the concentration of salts falls in blood, the hypothalamus stops ADH secretion. The walls of the collecting duct become less permeable to water. So, less water is reabsorbed, resulting in the excretion of dilute urine. As a result, the blood volume and blood pressure decrease.

Tidbit
ADH increases the blood volume. It results in an increase in blood pressure. ADH also causes the blood vessels to constrict or narrow. This constriction further increases blood pressure.

A person who cannot produce enough ADH due to pituitary damage constantly excretes a large volume of dilute urine. This disorder is known as *diabetes insipidus*. Such a person is in danger of becoming severely dehydrated with very low blood pressure.

2- Aldosterone

Aldosterone is the hormone responsible for regulating sodium and potassium levels of urine. It is produced by the **adrenal cortex** (the outer layer of adrenal gland present on the top of kidney).

Aldosterone is secreted when blood pressure drops or sodium levels in the blood are too low. It acts on the walls of the distal convoluted tubule and the collecting duct. It increases sodium ions reabsorption from the filtrate into the blood. Due to higher concentration of sodium ions in blood, water also moves from filtrate into the blood.

Tidbit
Aldosterone also promotes the excretion of potassium ions in the urine. It helps to prevent hyperkalaemia, a condition in which the blood potassium levels become too high and can lead to dangerous cardiac arrhythmias.

Two capillaries Beds in Kidneys
In kidney, two distinct capillary beds work in sequence to process blood.

1. **Glomerular capillaries** are located within the Bowman's capsule. These capillaries are designed for filtration. They operate under high blood pressure which forces water and small solutes out of the blood and into the nephron.
2. **Peritubular capillaries** surround the renal tubules. Their primary role is reabsorption and secretion. They operate under low hydrostatic pressure and pull essential nutrients (glucose, ions) and water back into the bloodstream from the tubules. They also secrete specific toxins into the tubule.

14.3- Disorders of Urinary Tract

A variety of disorders can affect the structure and function of kidneys and urinary tract. Common disorders include urinary tract infections, kidney stones, kidney

failure, and polycystic kidney disease.

14.3.1- Urinary Tract Infections

Urinary tract infections (UTIs) are usually caused by bacteria, most commonly *Escherichia coli*. These bacteria enter the urinary system through urethra and infect the bladder, ureters, or kidneys.

The symptoms of a UTI include a frequent and urgent need to urinate, pain or burning during urination, cloudy or strong-smelling urine, lower abdominal pain, and sometimes fever or chills. Treatment involves a course of antibiotics to kill the causative bacteria. Drinking plenty of water and urinating frequently can also help to flush out the bacteria and alleviate symptoms.

14.3.2- Kidney Stones

Urinary stones are hard, crystalline mineral materials that-stick together to form small "pebbles" within the kidney or urinary tract. They may stay in kidneys or travel out of the body through the urinary tract.

70% of kidney stone patients have stones of calcium oxalate. These stones are formed due to **hypercalciuria** (high calcium in urine) and **hyperoxaluria** (increased level of oxalate level in urine). 10% of patients may have stones of uric acid. **Hyperuricemia** (increased amount of uric acid in blood) is the cause of such stones. High concentration of cysteine and phosphates in urine also cause kidney stones. Continuous dehydration in the body increases the chances of kidney stone formation. Treatment of kidney stones usually involves lithotripsy i.e., breaking of stones. There are two methods of lithotripsy.

Tidbit

Doctors diagnose the nature of kidney stone by studying the urine test of patient.

When urine is acidic, the stone is of calcium oxalate.

When the urine is alkaline, the stone is of calcium phosphate.

When urine is persistently acidic, the stone is of uric acid type.

In **Extracorporeal Shockwave Lithotripsy (ECSWL)**, an instrument called **lithotripter** generates shockwaves from outside the patient's body. These shockwaves break the stone into small pieces. Most of pieces then pass out through urine.

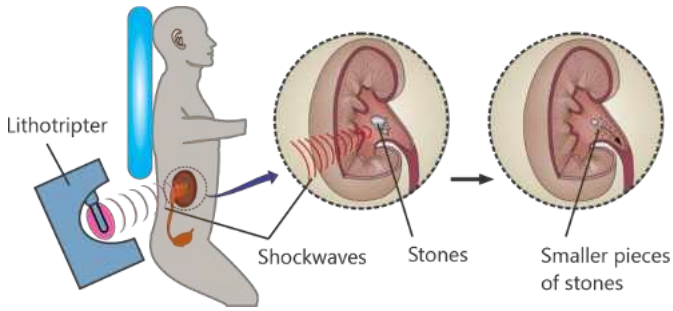


Figure 14.5: Extracorporeal shockwave lithotripsy

In case of larger stones, **Percutaneous Nephrolithotripsy** (PNL) is used. In this technique, a tube is inserted from the patient's back into the kidney. A small camera called **nephroscope** is inserted through the tube to visualize the stone. Ultrasound equipment is then inserted to break the stone. The stone pieces can be grasped with special equipment and pulled out from the kidney.

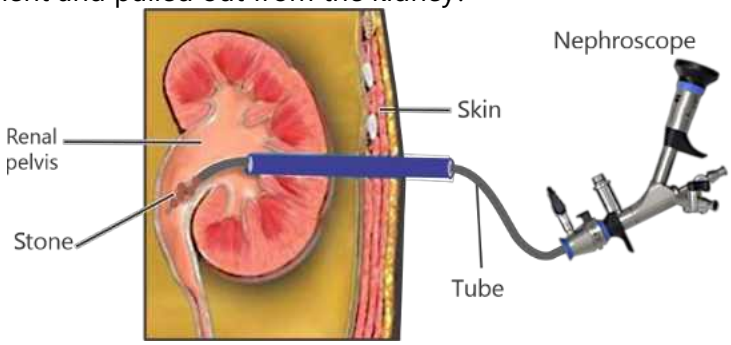


Figure 14.6: Percutaneous Nephrolithotripsy

14.3.3- Kidney Failure

Kidney/renal failure is a condition in which the kidneys lose their ability to filter wastes from the blood and regulate fluid and salt balance in the body. Chronic kidney failure develops over a longer period of time and is irreversible. It is often caused by underlying conditions like diabetes, high blood pressure, or kidney disease (chronic infection, inflammation of glomeruli etc.).

Acute kidney failure (also called kidney injury) occurs suddenly and often resolves with treatment. It may be due to kidney infection, blockage in urinary tract (due to kidney stones, bladder cancer, or enlarged prostate), certain medications such as NSAIDs, and blood clots in renal arteries or arterioles.

Renal dialysis is the treatment of renal failure in which the metabolic wastes are filtered from blood by artificial methods. There are two general types of renal dialysis i.e., haemodialysis & peritoneal dialysis.

1- Haemodialysis

In haemodialysis, the wastes are removed by circulating the blood outside the body through an equipment called dialyzer. This equipment is a type of filter and contains a semipermeable membrane (made of cellulose).

In this treatment, a catheter is inserted into an artery, usually in the arm. The blood is then circulated through dialyzer which also contains a special fluid called **dialysate**. As the blood passes through the dialyzer, the dialysate attracts the wastes, water, and salts to move out from blood, the filtrates passes through semipermeable membrane and dissolves in dialysate. The clean blood then returns to the body through a second catheter inserted in a vein.

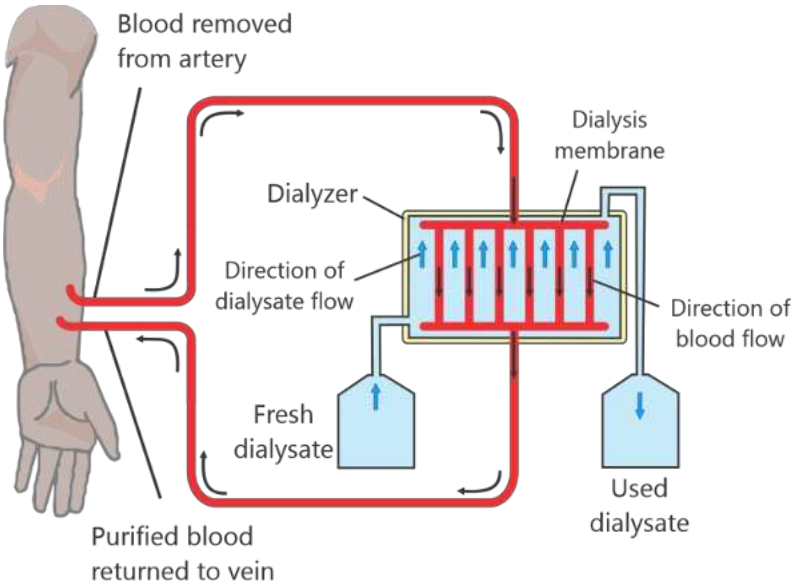


Figure 14.7: Haemodialysis

2- Peritoneal Dialysis

Peritoneal dialysis involves the use of peritoneal membrane that lines the abdominal cavity. In this process, 1.5 to 2 litre of dialysate is infused into the patient's abdomen through a catheter. The dialysate is left in abdomen for 4 to 6 hours. Waste products and excess fluid from the blood capillaries of peritoneum pass into the dialysate. Then, the dialysate is drained out of the abdomen and discarded. This process is repeated several times a day, to achieve adequate clearance of waste products and excess fluid.

Problems associated with dialysis

While dialysis can be life-saving for people with kidney failure, there are several problems associated with this treatment.

- Dialysis requires access to the bloodstream, which increases the risk of infections.
- Hypotension, or low blood pressure, is a common complication of haemodialysis.
- Muscle cramps can occur during dialysis due to changes in salt levels.
- Dialysis can cause anaemia, which can lead to fatigue and weakness.
- If too much fluid is removed too quickly, it can lead to dehydration or low blood pressure.
- Dialysis treatments can be emotionally challenging. Patients may feel anxious, depressed, or overwhelmed by the demands of the treatment.

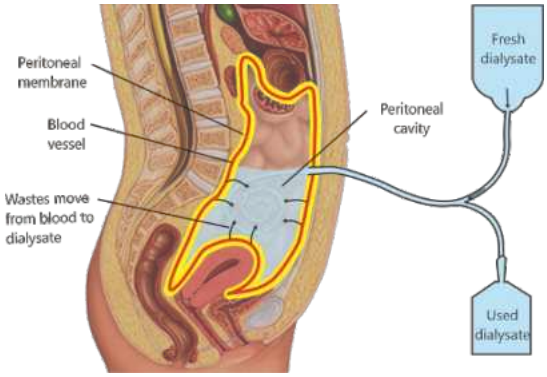


Figure 14.8: Peritoneal Dialysis

14.3.4- Kidney Transplant

Kidney transplant is a surgical procedure in which a person's non-functioning kidney is replaced with a healthy kidney from a donor. This treatment is offered to individuals with end-stage kidney failure. Kidney transplant process requires careful evaluation, matching, and monitoring to ensure the best results for both the recipient and the donor.

Potential Donors for Kidney Transplant

Related donor: A person who is genetically related to the recipient, such as a parent, sibling, or child can be the best donor of kidney.

Unrelated donor: The donor cannot be genetically related to the recipient, such as a spouse, friend, or co-worker. This type of donation is less common but can still be successful if the donor's and recipient's blood group and tissue type match.

Deceased donor: Many people register with hospitals for donation of their organs after their death. Their organs, including the kidneys, can be donated to individuals in need of a transplant. Deceased donor's kidneys are carefully matched to recipients based on blood group, tissue type, and immune system compatibility.

Tidbit

Kidney donation can be a life-saving gift for someone with end-stage kidney disease.

Donating a kidney does not have significant long-term health risks for the donor. Studies have shown that kidney donors generally experience no negative impact on their overall health or lifespan.

For Information

Pakistan's first kidney transplant centre was established in 1985 at the Sindh Institute of Urology and Transplantation (SIUT) in Karachi. It is one of the largest and most successful transplant centres in the world.

The Pakistan Kidney and Liver Institute & Research Center (PKLI&RC) was established in 2017. It is a tertiary-care hospital located in Lahore. It was established to address the massive national crisis of kidney and liver diseases, serving as a specialized center for medical care, transplantation, and research

Problems associated with kidney transplant

There are some potential complications and risks associated with kidney transplant. For example;

- The recipient's immune system may recognize the transplanted kidney as a foreign object and attack it, leading to organ rejection. This can occur even if the patient is taking immunosuppressant medications to prevent rejection.
- Patients with a suppressed immune system are at a higher risk of developing infections after a transplant.
- Recipients are given immunosuppressant medications to prevent organ rejection. Such medicines can have side effects such as high blood pressure, weight gain, and increased risk of infections and cancers.

EXERCISE

SECTION 1: MULTIPLE CHOICE QUESTIONS

1. Which one of these is not a part of single nephron?
   (a) Distal convoluted tubule (b) Collecting duct
   (c) Bowman's capsule (d) Loop of Henle
2. In a healthy person, glucose is present in blood but not in urine. This is because glucose molecules are;
   (a) Reabsorbed from the proximal convoluted tubule to blood
   (b) Oxidised in kidneys
   (c) Stored in kidneys
   (d) Too large to enter Bowman's capsule
3. The sizes of molecules present in Bowman's capsule are smaller than the sizes of molecules present in;
   (a) Afferent renal arteriole (b) Collecting duct
   (c) Loop of Henle (d) Proximal tubule
4. If a drug reduces mitochondrial activity in nephrons, which chemical will be present in increased amounts in the urine?
   (a) Ammonia (b) Glucose (c) Uric acid (d) Urea
5. The water content of blood is regulated by ADH. In which part of the nephron this regulation occurs?
   (a) Bowman's capsule (b) Proximal convoluted tubule
   (c) Loop of Henle (d) Collecting duct
6. In kidneys, glucose is reabsorbed into blood. Where does this reabsorption occur?
   (a) Bowman's capsule (b) Glomerulus
   (c) Proximal convoluted tubule (d) Collecting duct
7. In normal conditions, which one is completely reabsorbed from renal tubule into blood?

(a) Urea (b) Water (c) Salts (d) Glucose

8. If the loops of Henle are absent in all nephrons, what would happen?
   (a) No urine formation
   (b) Formation of urine with normal concentration
   (c) Formation of concentrated urine
   (d) Formation of dilute urine
9. In peritoneal dialysis, the lining of which part of the body acts as a filter?
   (a) Stomach (b) Intestine (c) Lungs (d) Abdomen
10. During which of these treatments, the patients are prescribed immunosuppressant drugs?
   (a) Lithotripsy (b) Haemodialysis
   (c) Kidney transplant (d)Peritoneal dialysis

SECTION 2: SHORT QUESTIONS

1. Justify the functioning of kidneys as both excretion and osmoregulation.
2. List the organs of the urinary system.
3. Define glomerular filtration.
4. Compare the function of glomerular capillaries and peritubular capillaries.
5. What are the three main processes involved with urine formation?
6. What are the causes of kidney failure?
7. Why do blood and dialysate flow in opposite direction?
8. Differentiate between:
   - Afferent and efferent arterioles
   - Chronic and acute renal failure
   - Haemodialysis and peritoneal dialysis
   - Renal cortex and renal medulla

SECTION 3: LONG QUESTIONS

1. Describe the structure of kidney and relate it with its function.
2. Explain the detailed structure of nephron.
3. Explain the main processes in kidney functioning.
4. Explain that concentration of urine is regulated by counter-curent and hormonal mechanisms.
5. Explain the causes and treatments of kidney stones.
6. Explain in detail the mechanism and problems related to dialysis.
7. Describe the principles and the problems associated with kidney transplant.

INQUISITIVE QUESTIONS

1. Hypothesize kidney stone by studying the urine test of relevant patients.
2. Describe the importance of kidney donation for the benefit of kidney failure patients.
3. Name the important kidney transplant centres in your city.

Chapter 15 HUMAN NERVOUS SYSTEM

STUDENTS'LEARNING OUTCOMES

After studying this chapter, the students will be able to:

- Recognize receptors as transducers sensitive to various stimuli.
- Trace the path of a message transmitted to the central nervous system for processing.
- Identify the three neurons (sensory, inter-, motor) involved in nervous transmission.
- Identify muscles and glands as the effectors.
- Annotate the detailed structure of a sensory neuron, associative and a motor neuron
- Relate the structure of neurons with functions.
- Differentiate between myelinated and non- myelinated neurons.
- Explain the function of the three types of neurons with the help of a reflex arc.
- Define nerve impulse.
- Describe the generation and transmission of nerve impulse.
- Name the factors responsible for the resting membrane potential of neuron.
- Evaluate from a graph the phenomena of polarization, depolarization, and hyperpolarization of membrane.
- Compare the velocities of nerve impulse in the axon membrane and in the synaptic cleft.
- Describe the role of local circuits in saltatory conduction of nerve impulse.
- Outline the structure of synapse.
- Explain synaptic transmission of nerve impulse.
- Classify neurotransmitters as inhibitory and excitatory and list some common examples.
- Identify the main components of the nervous system.
- Explain briefly the major parts functions of major divisions of the brain and their functions.
- Describe the architecture of human brain.
- Describe cranial and spinal nerves in man.
- Explain the structure, types and functions of the autonomic of autonomic nervous system.
- Explain the structure and functioning of the receptors for smell, taste and touch / pain.
- Define narcotic drugs as agents that interact with the normal nervous activity.
- Compare the use and abuse of drugs with respect to heroine, Cannabis, nicotine, alcohol and inhalants like nail polish remover and glue.
- Explain the terms; drug addiction and drug tolerance with reference to caffeine and nicotine and their adverse effects.
- Associate the effects of drug addiction and tolerance with the functioning of the nervous system.
- Describe the way how pain medicines can reduce or numb pain in the human body.
- Discuss that certain pain medications are addictive.

- Classify nervous disorders into vascular, infectious, structural, functional and degenerative disorders.
- Describe the causes, symptoms and treatment of one type of each category of disorders outlined above.
- Explain the principles of the important diagnostic tests for nervous disorders i.e., EEG, CT scan and MRI.

To survive, every animal must stay connected to the world around it. They need to sense and respond to changes in environment. The activities of their organs and systems are also coordinated. For example; the heart rate, breathing rate, digestion, and many other functions are coordinated. Due to such coordination, every function is regulated according to the needs of the body. In humans and other complex animals, there are two types of coordination i.e.,

Tidbit
Think of your body as a large, complex city. For the city to run smoothly, the traffic lights, emergency services, and power plants must all talk to each other. This "talking" is what we call **coordination**.

i- Nervous coordination controlled by the nervous system
ii- Chemical coordination controlled by the endocrine system

This chapters deals with details of the human nervous system.

15.1- Nervous Coordination

In nervous coordination, the nervous system controls coordination with external environment and among the different parts of the body. Nervous coordination is done by means of electrical signals.

15.1.1- Basic Elements of Nervous Coordination

For homeostasis, the nervous system is the most important because it regulates other systems and also maintains homeostasis by itself. You have studied the basic elements of homeostasis. Here, you will study them with reference to the nervous system.

1. **Receptors:** These are organs, tissues or cells – such as eyes, ears, or skin – that detect stimuli (the changes in the external and internal environment). They act as transducers. When they receive stimuli, they generate nerve impulses in **sensory neurons**. For processing the information, the sensory neurons carry nerve impulse to central nervous system.

Tidbit
Receptors act as transducers because they convert one form of energy into another form e.g., rod and cone cells in the retina of eye convert the light energy into nerve impulse (electrochemical energy).

2. **Central nervous system:** It is the control centre of nervous coordination. It consists of brain and spinal cord. These parts are made of **associative neurons** (also called **inter-neurons**). They receive the nerve impulses from sensory neurons and process them. After processing, they send nerve impulses to effectors via **motor neurons**.
3. **Effectors:** These are the parts which produce responses on receiving nerve impulse sent by the central nervous system via motor neurons. Muscles and glands act as effectors. Muscles produce response by contracting while glands produce secretion.

So, in nervous coordination, the path of a message (nerve impulse) consists of receptors, sensory neurons, central nervous system (inter-neurons), motor neurons, and effectors. In this pathway, the neurons act as the functional units.

15.2- Neurons

The function of the nervous system (responding to the environment and coordinating body activities) is actually carried out by specialized celled neurons (also called nerve cells). A neuron is the structural and functional unit of nervous system.

15.2.1- Structure of Neuron

There are many variations in the structure of neurons. But all neurons have the three basic components i.e., a cell body, dendrites and an axon.

The **cell body** is an enlarged region of the neuron cell. The nucleus of neuron and most of its organelles are located in cell body. The cytoplasm in cell body is characterised by the presence of **Nissl's granules**. These are group of ribosomes and rough endoplasmic reticulum.

Dendrites are membrane-bounded extensions of cell body. Dendrites receive information from other neurons or other cells. They convert the information into nerve impulses and carry it toward cell body. From cell body, the impulses are conducted away along an axon. Motor and inter-neurons receive impulses from many different sources simultaneously. That is why, they possess highly branched dendrites.

Axon is a long and membrane-bounded extension of cell body. It transmits impulses away from cell body. A neuron may have a single axon or branching axons. The end of an axon is called the **axon terminal**. It may contact and communicate with a muscle cell, a gland cell, or another neuron.

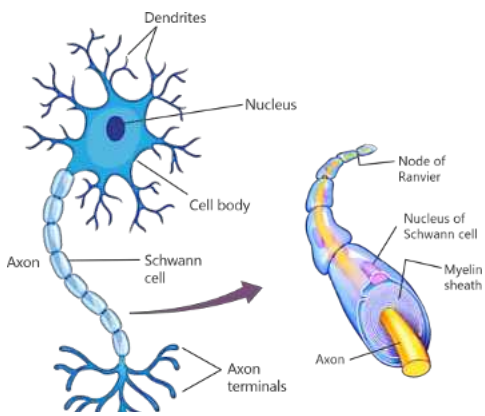


Figure 15.1: Structure of a model neuron

Tidbit
An axon can be quite long: the axons controlling the muscles in your feet are more than a meter long, and the axons that extend from the skull to the pelvis in a giraffe are about three meters long!

Supporting Cells – The Neuroglia

In the nervous system, there are special cells which support neurons. These supporting cells are called **neuroglia** or **glial cells**. They supply nutrients to neurons, remove wastes, and provide immunity. Two important glial cells are Schwann cells and oligodendrocytes. They are wrapped around the axons of many neurons. Here, they make myelin sheaths (sheaths of multiple membrane layers) around the axons. **Schwann cells** produce myelin sheaths around the axons in the peripheral nervous system. **Oligodendrocytes** produce myelin sheath in central nervous system.

Myelin sheath is an insulating covering. It helps in the fast transmission of nerve impulse along the axons. The neurons which have myelin sheaths around axons are called **myelinated**, and those that do not have such covering are called **non-myelinated**. In the CNS, myelinated axons make **white matter**, and the non-myelinated dendrites and cell bodies make **grey matter**. In the peripheral nervous system, both myelinated and non-myelinated axons are bundled together to form nerves. In myelinated neurons, the myelin sheath is interrupted at intervals of 1 to 2 mm by small gaps (non-myelinated parts). These non-myelinated interruptions are called **nodes of Ranvier**.

15.2.2- Types of Neurons

Sensory neurons conduct impulses from receptors to central nervous system. Their cell bodies are located outside of the CNS in the form of clusters called **ganglia**. Their cell body gives rise to a dendrite on one side and an axon at the other. Sensory neurons are unipolar. It means that they have only one long dendrite. The terminal end of dendrite has many smaller branches, which are connected to the receptors.

Motor neurons conduct impulses away from central nervous system. They have a cell body on one end, a long axon and many dendrites. Their cell bodies are located in the CNS. They are multipolar. It means that they have many dendrites that branch out into many smaller dendrites. Their dendrites make contact with other neurons in brain and spinal cord.

Inter-neurons (associative neurons) occur entirely within the CNS. They receive impulses from sensory neurons or other inter-neurons and transmit impulses to motor neurons or other inter-neurons. Some inter-neurons have short axons and form circuits with nearby neurons to analyse small information. Other inter-neurons have long axons and connect with other inter-neurons in different regions of the brain and spinal cord.

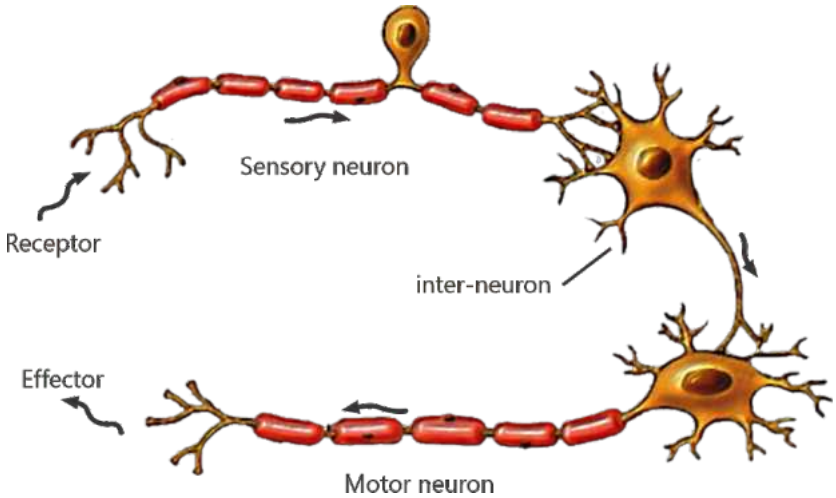


Figure 15.2: Types of neurons

15.2.3- Reflex Arc

In nervous system, neurons follow specific neural pathways (arcs) to receive, transmit and process impulses. Neural pathways may compose of only a few neurons, or may be more complex networks. The reflex arc is the simplest neural pathway. It controls reflex action i.e., immediate, automatic and involuntary responses to external and internal stimuli.

The Knee-Jerk Reflex

Knee-jerk is a reflex that occurs when someone hits the tendon below the knee (the patellar tendon) with a hammer. Tapping on that tendon acts as a stimulus. This stimulus (tapping) is sensed by a receptor present in the quadriceps muscle of thigh. The receptor generates a nerve impulse in a sensory neuron. The sensory neuron carries this impulse travels to spinal cord. Here, the sensory neuron transfers the impulse to a motor neuron without involving an inter-neuron. The motor neuron carries the impulse to the effector (quadriceps muscle) which contracts. It leads to extension of the lower leg at the knee (i.e., the lower leg kicks forward).

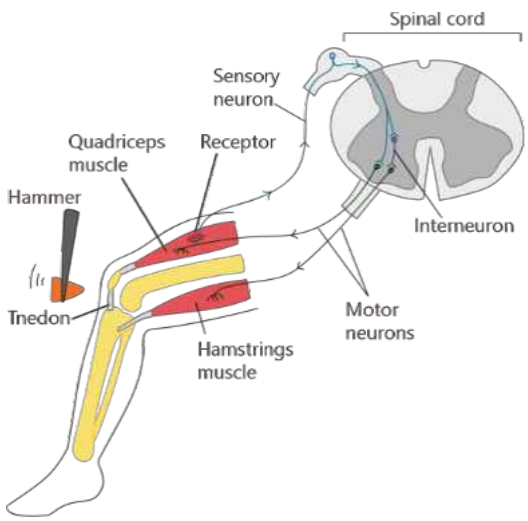


Figure 15.3: Knee-jerk reflex

In the meantime, sensory neuron also activates an inter-neuron in spinal cord. The inter-neuron releases chemicals which inhibit the motor neurons that connect to the hamstrings muscle. It results in the relaxation of hamstrings during the extension of the lower leg.

15.3- Nerve Impulse

A nerve impulse is defined as a wave of electrochemical changes that travels along the length of neuron. All cells, including neurons, have electrical charges on their inner and outer sides. The inner side of membrane (towards cytoplasm) is electrically negative while the outer side (towards extracellular fluid) is positive. This difference in the electrical charges across cell membrane is called **membrane potential**.

These membrane potentials are due to different concentrations of positive and negative ions and due to the movement of ions across cell membrane. In most cells, the inward and outward movement of ions through the membrane is constant. So, the net negative charge on the inner side remains constant. However, the cell membranes of muscle and neurons can change the rates of ions movements. So, their membrane potentials also change.

15.3.1- Generation and Transmission of Nerve Impulse

1- Before Nerve Impulse - Resting Membrane Potential

A resting neuron (not conducting a signal) is said to be **polarized**. Its inner side has negative while outside has positive charge. This membrane potential of a polarized neuron is called Resting Membrane Potential (RMP) and it is about –70 millivolts (–70 mV). The negative sign indicates that the inside of the cell is negative with respect to the outside. In resting neurons, RMP is established by the following factors:

1. Large negatively charged molecules e.g., proteins and nucleic acids are more abundant inside the cell and cannot diffuse out. These molecules are called **fixed anions**.

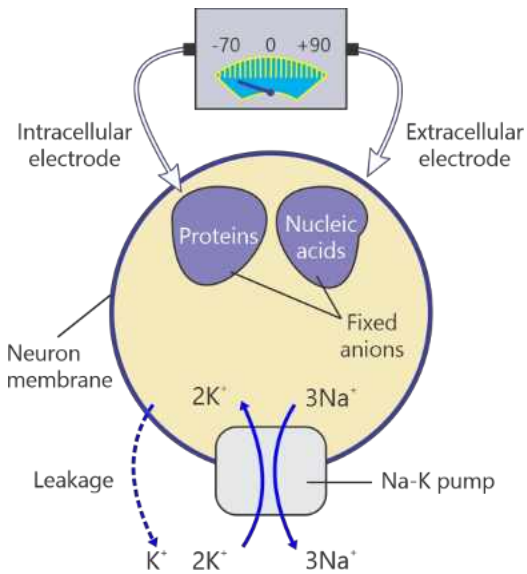


Figure 15.4: Factors responsible for RMP

2. The concentration of K^+ is 30 times greater inside the neuron than outside. While, the concentration of Na^+ is 10 times greater outside than inside the cell. Sodium-potassium pumps present in neuron membranes constantly move these ions against their concentration gradient by the expenditure of ATP. For every two K^+ that are actively transported inward, three Na^+ are pumped out. So inside becomes more negative than outside.
3. Some K^+ ions also leak out of the cell through K^+ gates of membrane.

For Information
This exit of K^+ and the retention of negatively charged proteins are the major factors for establishing net negative charge inside and a positive charge outside i.e., RMP.

2- Generation of Nerve Impulse - Active Membrane Potential

When a neuron becomes active (generates nerve impulse), the membrane potential at a region of neuron is reversed i.e., the inside becomes positive than the outside. It is called Active Membrane Potential (or action potential).

The following events happen for producing action potential.

- **Depolarization:** The Na^+ gates of membrane open and Na^+ ions rush into the cell. This sudden inward movement of Na^+ ions reduces the negative charge on the inside. The electrical potential of the membrane changes from -70 mV towards zero and then reaches to 50 mV. This reversal of polarity across two sides of membrane is called depolarization. This depolarization produces action potential on a short region of neuron for about one millisecond.
- **Repolarization**: After a short period of depolarization, Na^+ gates close and the K^+ gates open. So, more K^+ ions diffuse out. It makes the inside of cell negative again. It is called repolarization of membrane (negative inside and positive outside).
- **Hyperpolarization:** The repolarization carries the membrane potential to a value slightly more negative than the resting potential for a brief period. This is called hyperpolarization**.** It is due to the slight delay in closing all the K^+ gates. During hyperpolarization, the neuron cannot generate another action potential and period is called the **refractory period**.
- **Return to Original RMP:** After a short refractory period, original ionic distribution is restored by sodium-potassium pump and the membrane returns to its RMP i.e., -70mV. In this way, the region of neuron prepares itself for the next action potential.

Tidbit
The propagation of action potentials is similar to the action when people in a stadium perform the "wave": Individuals stay in place as they stand up (depolarize), raise their hands (action potential), and sit down again (repolarize).

3- Propagation of Action Potentials

In reality, action potentials (impulses) do not really travel along the axon. Rather, they are reproduced at adjacent regions along the axon. An action potential in a region of axon serves as a depolarization stimulus for the next region. So, the next region produces its own action potential. Meanwhile, the previous region repolarizes back to the RMP.

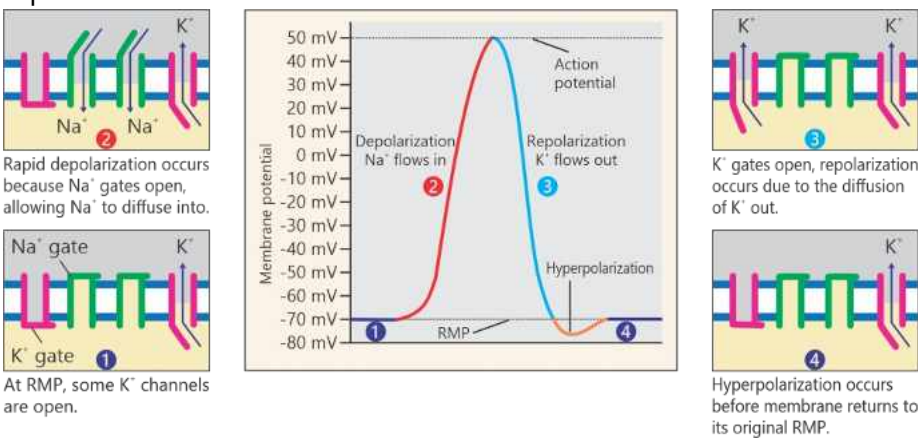


Figure 15.5: Generation of action potential

15.3.2- Velocities of Nerve Impulse

The strength (amplitude) of action potentials remains constant along the length of axon. The action potential at the end of axon is as strong as the first action potential. However, the velocity of nerve impulse i.e., speed of conduction of action potential may vary. The velocity is greater if the diameter of the axon is large or if the axon is myelinated.

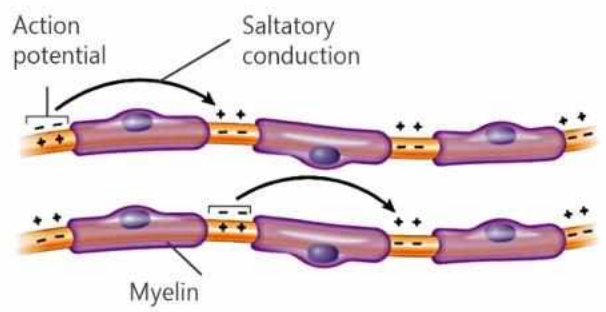


Figure 15.6: Saltatory conduction in myelinated axon

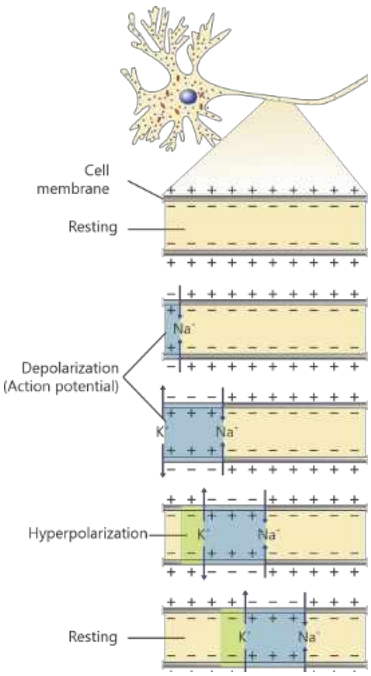


Figure 15.5: Propagation of action potential

In myelinated axons, action potentials are only produced at the nodes of Ranvier. These action potential jump from one node of Ranvier to another, skipping the myelinated regions of membrane. It is called as **saltatory conduction** of nerve impulse. It is up to 50 times faster than the nerve impulse through the fastest non-myelinated axons. In human, the non-myelinated neurons conduct impulses at speed of 1 to 3 metres per second. While, the myelinated neurons conduct at speeds of up to 120 meters per second.

15.4- Synapse

The junction between axon terminal of one neuron and the dendrites of other neurons, or effector cells (of muscle or gland) is called a synapse. At a synapse, the two neurons or a neuron and a cell are not in direct contact. Rather, there is a fluid-filled gap between them, called a **synaptic cleft**. The neuron whose axon transmits action potentials towards synapse is the **presynaptic** neuron. The neuron or cell on the other side of the synapse is the **postsynaptic** cell.

For Information
The synapse between a motor neuron and a muscle fibre is also called **neuro-muscular junction.**

15.4.1- Mechanism of Synaptic Transmission

The axon terminal of presynaptic neuron has several **synaptic knobs**. These knobs contain numerous **synaptic vesicles** which are packed with chemicals called **neurotransmitters**. When an action potential on a presynaptic neuron reaches a synapse, its information is transmitted to postsynaptic neuron or effector cell. The movement of action potential across synapse is called **synaptic transmission**. In happens in the following way:

1. When action potential arrives at synaptic knobs, calcium gates in presynaptic membrane open.
2. Calcium ions present in synaptic cleft diffuse rapidly into presynaptic axon.
3. Due to the diffusion of calcium ions into presynaptic axon, its synaptic vesicles fuse with its membrane. This fusion causes the release of neurotransmitters from vesicles by exocytosis.
4. The released neurotransmitters diffuse rapidly in the cleft and bind to postsynaptic membrane at receptor proteins.

For Information
The higher the frequency of action potentials in the presynaptic axon, the more neurotransmitters are released.

5. The binding of neurotransmitters acts as a massage for postsynaptic membrane. Its Na^+ gates open and an action potential is generated in postsynaptic membrane.
6. After their action, the neurotransmitters do not remain in the synaptic cleft indefinitely. At many synapses, the used neurotransmitters are reabsorbed by the presynaptic neuron. At other synapses, they are broken down by enzymes.

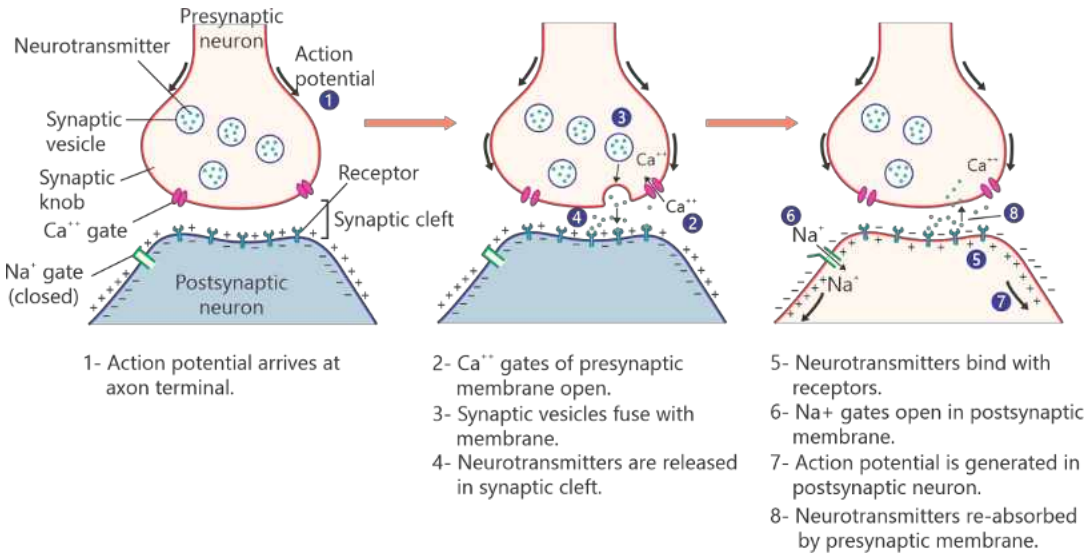


Figure 15.7: Structure of synapse and mechanism of synaptic transmission

15.4.2- Types of Neurotransmitters

i- Excitatory Neurotransmitters

Excitatory neurotransmitters generate nerve impulse (action potential) in postsynaptic neurons by increasing membrane permeability to Na^+. For example,

- **Acetylcholine** acts between motor neurons and skeletal muscle fibres, and between many neurons.
- **Glutamate** acts in the CNS.
- **Biogenic amines** are excitatory neurotransmitters. They also function as hormones. These include;
  - i- **Norepinephrine** is made in CNS and sympathetic nerves. It regulates sleep patterns, focus and alertness.

For Information

Degeneration of dopamine-releasing neurons produces the muscle tremors of Parkinson's disease.

Insufficient activity of serotonin-releasing neurons may cause depression. Many antidepressant drugs block the elimination of serotonin from the synaptic cleft. So, serotonin remains in the synaptic cleft and reduces depression.

ii- **Epinephrine** is released in adrenal glands and the brainstem. It plays a role in sleep. It also enables to become and stay alert.

iii- **Dopamine** is a neurotransmitter in brain. It controls body movements, pleasures related to motivation and also emotional stimulation.

iv- **Serotonin** is produced by the neurons in intestine (approximately 90%), and CNS (10%). It regulates appetite, sleep, memory and learning, temperature, mood, behaviour, muscle contraction etc.

ii. Inhibitory Neurotransmitters

These neurotransmitters decrease the chance of transmission of nerve impulse to the postsynaptic neuron. When they bind to the receptors on postsynaptic neuron, channels for chloride ions open instead of opening sodium channels (which would excite the cell). As a result, chloride ions diffuse into neuron and make the inside of the membrane even more negative than RPM. This hyperpolarization causes inhibitory effect. Examples of inhibitory neurotransmitters include:

- **Gamma - aminobutyric acid (GABA)** is the chief inhibitory neurotransmitter in CNS. It reduces the neural excitability throughout the nervous system. It helps to control and regulate various physiological and behavioural processes. It reduces anxiety, stress, and fear, and promotes relaxation and sleep.

For Information
Many sedative drugs enhance the binding of GABA to its receptors and so increase the effectiveness of GABA at the synapse.

- **Glycine** is an inhibitory neurotransmitter especially in the spinal cord, brainstem, and retina.

Endorphins are inhibitory neurotransmitters released by neurons in brain. They reduce the release of excitatory neurotransmitters responsible for transmitting pain messages. In this way, they block the pain signals to reach the concerned part of the brain.

For Information
Opium and its derivatives, morphine and heroin, have an analgesic (pain-reducing) effect because they are similar in chemical structure to endorphins. So, they easily bind to the receptors of endorphins.

Effect of nerve gas on neurotransmitter acetylcholine
The excitatory neurotransmitter acetylcholine is released by motor neurons at neuromuscular junctions. It stimulates the skeletal muscles to contract. Then, it is broken down by acetylcholinesterase enzyme. A nerve gas (e.g., sarin, tabun, soman) is a highly toxic chemical that inhibits this enzyme. This inhibition leads to the accumulation of acetylcholine, causing overstimulation of skeletal muscle and cause muscles to remain contracted. If the diaphragm muscle remains contracted, the person is not be able to breathe. Other effects may include muscle twitching, convulsions, respiratory distress, and ultimately death.

15.5- Basic Organization of Human Nervous System

The human nervous system consists of central nervous system (CNS) and peripheral nervous system (PNS). The **CNS** consists of brain and spinal cord. The **PNS** consists of cranial and spinal nerves which arise from brain and spinal cord respectively. The nerves make sensory and motor pathways. There are two motor pathways: (i) voluntary (somatic) nervous system which controls voluntary (skeletal) muscles, and (ii) autonomic nervous system which controls involuntary muscles and glands. The autonomic system consists of sympathetic and parasympathetic nervous systems.

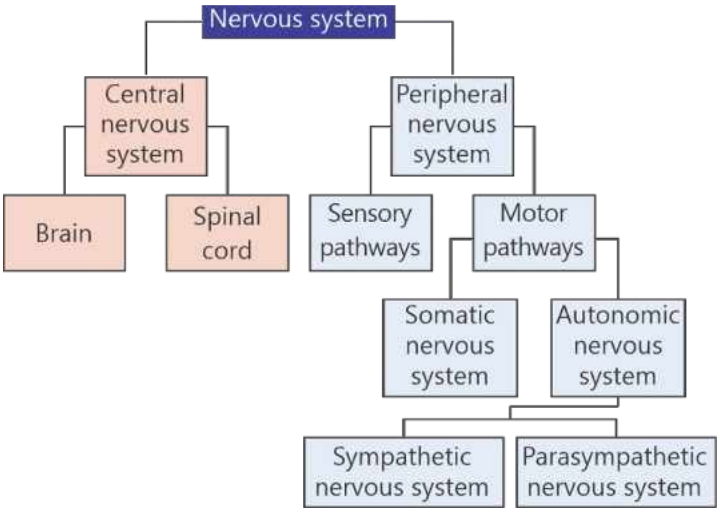


Figure 15.8: Basic organization of nervous system

15.5.1- Central Nervous System – Brain & Spinal Cord

The CNS (brain and spinal cord) is made of inter-neurons and neuroglia. Brain is enclosed within the cranium while spinal cord is enclosed within vertebral column. Both are covered with protective **meninges** which consists of three layers of membranes;

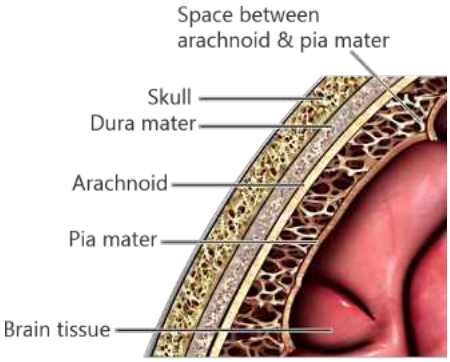


Figure 15.9: The meninges of brain

i- The outermost thick layer (below skull and vertebrae) is called **dura mater**. It contains larger blood vessels.

ii- The second layer (below dura mater) is thin and is called **arachnoid**. It acts as a barrier between the cerebrospinal fluid present below it and the blood in the dura mater.

iii- The third layer is very delicate membrane called **pia mater**. It adheres to the surfaces of brain and spinal and has blood capillaries that nourish the brain and spinal cord.

Structure of Brain

The average adult human brain weighs 1.4 kg (about 2 percent of total body weight). It contains about 100 billion inter-neurons. Each neuron has synapses with several thousand other neurons.

For Information
In Grade X, you have studied the traditional divisions of brain structure i.e., forebrain, midbrain, and hindbrain. Here, you would study the structure of brain according to the latest defined divisions.

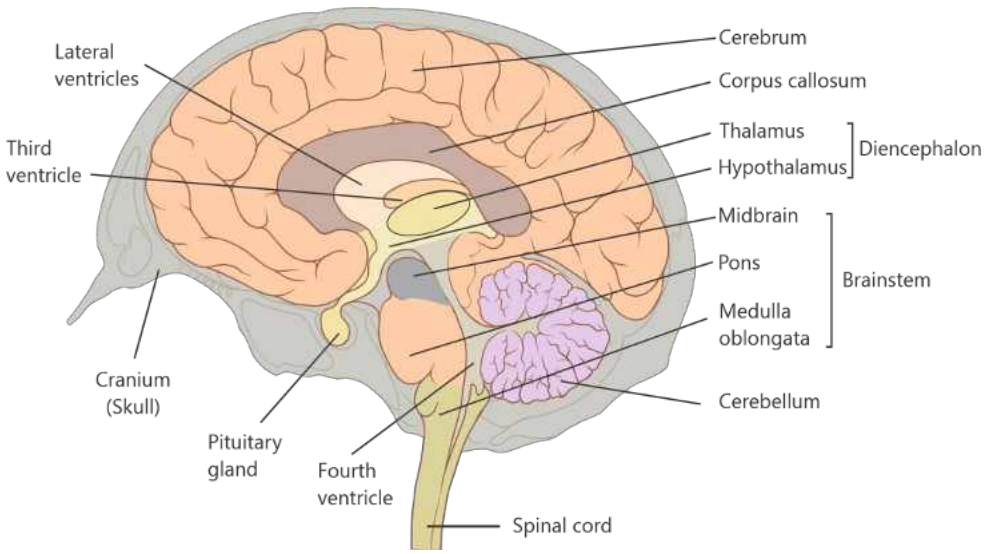


Figure 15.10: Major parts of human brain

The human brain consists of the following major parts.

1- Cerebrum

It is the largest portion of human brain. Its outer layer is highly folded. It is divided by a deep groove (fissure) into two parts called left and right cerebral hemispheres. At the base of deep groove, both cerebral hemispheres are connected by the **corpus callosum** (a band of axons). Each hemisphere receives sensory information from the opposite side of the body and sends motor commands to that side.

Tidbit
Damage to one hemisphere results in a loss of sensation and paralysis on the opposite side of body.

Each cerebral hemisphere is divided into four lobes:

i- The **frontal** lobes are associated with executive functions including self-control, planning, reasoning, and abstract thought.
ii- The **parietal** lobes process sensory information, give awareness of position of the body and the environment, control attention, plan and execute motor movements.
iii- The **temporal** lobes are involved in processing sensory input for appropriate retention of visual memory, language comprehension, and emotions.
iv- The **occipital** lobes control vision.

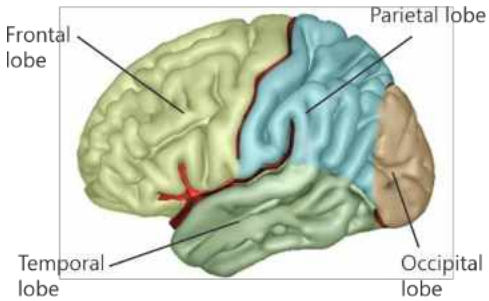


Figure 15.11: Four lobes of cerebrum

Tidbit
Ridges in cerebral hemispheres are called **gyri** (singular, gyrus). Gyri are separated by grooves. A shallow groove is called **sulcus** (plural, sulci) and a deep groove is called **fissure**.

Cerebral Cortex: It is the folded outer layer of cerebrum. It is made of grey matter (containing cell bodies and non-myelinated axons). It is densely packed with inter-neurons (about 10% of all neurons in brain). It contains the following areas.

i- The **motor areas** send impulses to the voluntary (skeletal) muscles.
ii- The **sensory areas** receive information from different parts. A specialized area called **auditory cortex** within the temporal lobe deals with different sound frequencies. The **visual cortex** in the occipital lobe deals with information from the eye.

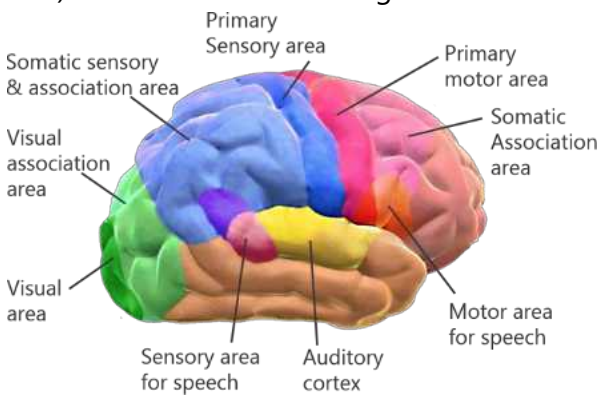


Figure 15.12: Sensory, motor and association areas

iii- **Association areas** of the cortex are the sites of higher mental activities. Some functions are associated with one side, such as language in the left and visual-spatial ability in the right.

Below cerebral cortex is the white matter (composed of myelinated axons). It links specific regions of the cortex with each other and with other neural centres.

2- Brainstem

The cerebrum is connected to the spinal cord through brainstem. This region lies below the cerebrum and consists of three main divisions:

i- **Midbrain**: It relays visual and auditory information. It also contains **reticular formation**, which connects hindbrain with forebrain.

ii- **Pons:** It transmits impulses between the cerebral hemispheres and cerebellum.

iii- **Medulla oblongata:** It serves as both a relay centre and a control centre for heart rate, respiration rate, and other homeostatic activities.

3- Diencephalon

It is present deep within the brain's center, above the brainstem and beneath the cerebrum (specifically between the cerebral hemispheres). It consists two major parts:

i- **Thalamus** is a relay centre between sensory information and the cerebrum.

ii- **Hypothalamus** integrates the visceral activities. It regulates body temperature, hunger and satiety, thirst, and various emotional states. It also controls the pituitary gland, which in turn regulates many of the other endocrine glands.

Tidbit
The pineal gland is located where the two halves of the thalamus join. It produces a hormone melatonin which controls sleep patterns.

Limbic system

It is a group of linked structures. It lies on both sides of the thalamus, just under the cerebrum. It includes the hypothalamus, the amygdala, the hippocampus, and several other nearby areas. The **amygdalae** (singular: amygdala) are two almond-shaped structures present deep within the temporal lobes. They control feeling and emotions of love, hate, anger, fear, rage and sexual arousal. The **hippocampus** consists of two "horns" that curve back from the amygdala. It plays role in the consolidation of information from short-term memory to long-term memory.

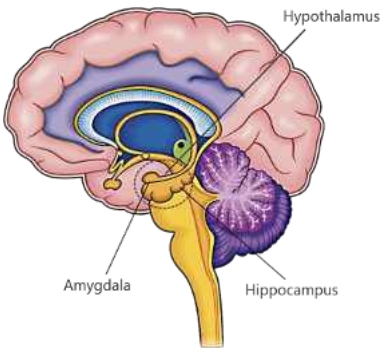


Figure 15.13: Limbic system

4- Cerebellum

It is the second major structure in brain. It lies below and behind the cerebral hemispheres. It coordinates muscle actions. It consists of a tightly folded layer of cortex, with white matter underneath. It receives sensory impulses from muscles, tendons, joints, eyes, and ears. It also receives information from other brain centres. It processes

information about body position and controls posture. The cerebellum coordinates rapid and ongoing movements. It acts with the brainstem and with the cerebral cortex to coordinate skeletal muscles.

Ventricles of Brain

The brain and spinal cord are hollow structure. It means that they contain fluid-filled cavities called ventricles. There are four ventricles in human brain. There are two large **lateral ventricles** in cerebrum. The **third ventricle** is in diencephalon between the right and left thalamus. The **fourth ventricle** is located between the cerebellum and the brainstem, and it is continuous with the central canal of spinal cord. The main function of ventricles is the production and circulation of cerebrospinal fluid.

Structure of Spinal Cord

The spinal cord is a cable of neurons extending from the medulla oblongata of brain down through the backbone. It is enclosed and protected by the vertebral column and layers of meninges. Like brain, the neurons and neuroglia of spinal cord make grey and white matter, but the arrangement of grey and white matter in spinal cord is opposite to that of brain.

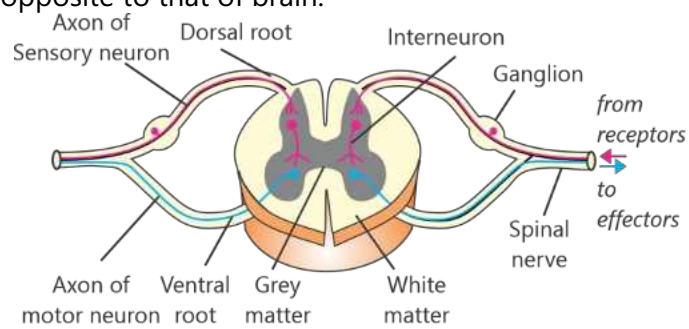


Figure 15.14: Structure of spinal cord and Spinal nerves

Tidbit

The diameter of spinal cord is between 0.25 to 0.5 inch. It is wider in cervical and lumbar regions. In cervical region, it deals with sensory input and motor output for arms and trunk. In lumbar region, it handles sensory input and motor output for legs.

There are two zones inside spinal cord. The inner H-shaped or butterfly-shaped zone is made of grey matter. It contains cell bodies of inter-neurons and motor neurons. It also contains neuroglia cells and non-myelinated axons. The outer zone is made of white matter. It contains myelinated axons. The inner zone also contains a central fluid-filled canal. It is a continuation of the fourth ventricle of brain. Spinal cord acts as information highway on which messages are transmitted between body and the brain.

15.5.2- Peripheral Nervous System – Cranial & Spinal Nerves

The peripheral nervous system consists of nerves and ganglia. Nerves are cable-like collections of axons. Ganglia are aggregations of cell bodies located outside CNS. The nerves which arise from brain are called cranial nerves while those which arise from

spinal nerves are called spinal nerves

Cranial Nerves

There are 12 pairs of cranial nerves. The first two pairs of cranial nerves (olfactory nerve and optic nerve) arise from cerebrum. The other 10 cranial nerve pairs arise from the brainstem. Some of cranial nerves are sensory, some are motor and others are mixed nerves (contain axons of sensory and motor neurons).

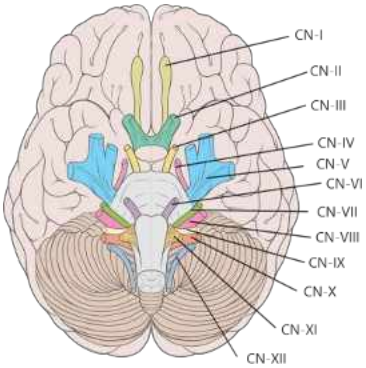


Figure 15.15: Cranial nerves

Table: Twelve Pairs of Cranial Nerves & their Functions

Cranial Nerve	Nature	Major Function
I	Sensory	Sense of smell
II	Sensory	Sense of vision
III	Motor	Movement of eye
IV	Motor	Movement of the superior oblique muscle of eye
V	Mixed	Sensation in the face; control of the muscles involved in chewing
VI	Motor	Movement of the lateral muscles of eye which are responsible for outward gaze
VII	Mixed	Facial expressions and the sense of taste in the front of the tongue.
VIII	Sensory	Hearing and balance
IX	Mixed	Sense of taste in the back of tongue; movement of the muscles involved in swallowing
X	Mixed	Controls many organs, including heart, lungs, and digestive system
XI	Motor	Controls the muscles in the neck and shoulders
XII	Motor	Controls the muscles of the tongue

Spinal Nerves

There are thirty-one pairs of spinal nerves. All spinal nerves are mixed. They transmit sensory and motor information between the spinal cord and the rest of the body.

Each spinal nerve emerges from the spinal cord by two short branches or roots. The dorsal root contains the axons and dendrites of sensory neurons. The ventral root contains the axons of motor neurons. The two roots join just before a spinal nerve leaves the vertebral column. The cell bodies of sensory neurons are grouped together outside spinal cord in the dorsal root ganglia. The cell bodies of somatic motor neurons are located within the spinal cord and so are not located in ganglia. The 31 pairs of spinal nerves include;

- 8 pairs of cervical nerves
- 12 pairs of thoracic nerves
- 5 pairs of lumbar nerves
- 5 pairs of sacral nerves
- 1 pair of coccygeal nerves

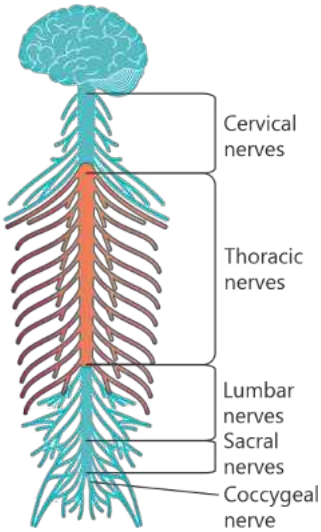


Figure 15.16: Spinal nerves

15.5.3- Divisions of the PNS

The PNS makes two types of pathways or divisions i.e., sensory pathways and motor pathways. The sensory pathways contain the sensory neurons of spinal and cranial nerves that connect the receptors of body to CNS. The motor pathways contain the motor neurons of spinal and cranial nerves that connect the CNS the effectors of body. The motor pathways make of two systems i.e., the somatic nervous system and the autonomic nervous system.

1- Somatic Nervous System

It consists of those motor neurons of spinal and cranial nerves which control skeletal muscles. The somatic system is voluntary. It means that skeletal muscles can be moved at will. For reflex action, the somatic system operates without conscious control.

2- Autonomic Nervous System

It consists of those motor neurons of spinal and cranial nerves which control smooth muscles and glands. The autonomic system is involuntary and it controls internal body conditions by regulating smooth muscles. For example, it controls respiration, heartbeat, digestion, and other functions. The autonomic system has two subdivisions i.e., the sympathetic division and the parasympathetic division.

a- Sympathetic Nervous System

It prepares the body under physical or emotional stress. The motor neurons of sympathetic nervous system originate from the thoracic and lumbar regions of spinal

cord. The axons of these neurons reach different visceral organs (smooth muscles and glands). When the sympathetic nervous system is activated, its motor neurons stimulate various smooth muscles and glands to prepare the body for fight or flight. For example, the heart beats faster and stronger, blood glucose concentration increases, blood flow is diverted to the muscles and heart, and the bronchioles dilate.

b- Parasympathetic Nervous System

This system antagonises the response of sympathetic nervous system after the threat or stress has passed. The motor neurons of parasympathetic nervous system originate from the brain and sacral regions of spinal cord. Many of these motor neurons are in vagus (the tenth cranial) nerve. These motor neurons reach the internal organs. These neurons bring the internal organs to their normal conditions. For example, heart beat slows down, blood flow to heart and skeletal muscles decreases, digestive organs increase secretion and other activities, and so on.

Table: Effects of Autonomic Nervous System on Body Parts

Body Parts	Effect of Sympathetic Nervous System	Effect of Parasympathetic Nervous System
Pupil of eye	Dilation	Constriction
Salivary glands	Decreased secretion	Increased secretion
Gastric glands	Inhibition of secretion	Stimulation of secretion
Liver	Stimulation of glucose secretion	Inhibition of glucose secretion
Digestive tract	Decreased motility	Increased motility
Urinary bladder	Relaxation	Contraction
Heart muscle	Increased rate and strength	Decreased rate
Bronchioles	Dilation	Constriction
Blood vessels in muscles	Dilation	No effect
Blood vessels in skin	Constriction	No effect

15.6- Receptors in Human Body

You know that receptors are the organs, tissues, cells or parts of cells that detect changes (stimuli) in external or internal environment and create nerve impulses in the associated sensory neurons. We will discuss the receptors for taste, smell, touch, and pain.

15.6.1- Receptors of Taste

Special chemoreceptors called taste buds sense the taste. There are about 10,000 taste buds. Most of them are embedded between bumps called papillae on the tongue. Some taste buds are present in the throat and on the roof of oral cavity. A taste bud is bulb-shaped and is made up of receptor cells with hair-like structures and basal cells (for making new receptor cells). A taste bud opens out into the mouth through a taste pore.

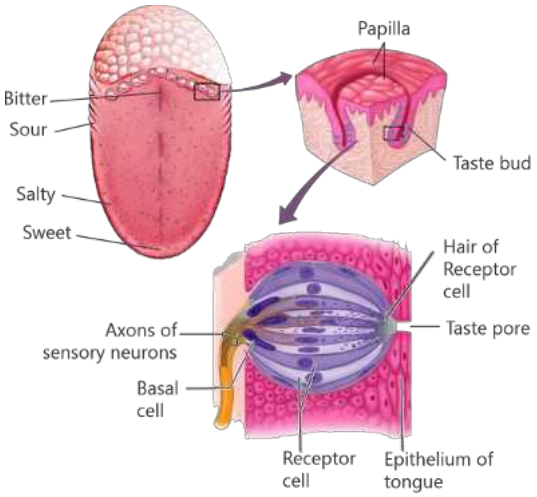


Figure 15.17: Receptors of taste

For Information
The simplest receptors are free nerve endings that respond various stimuli e.g., changes in temperature, chemicals in extracellular fluid.

For Information
The effect of smell on the sense of taste can easily be demonstrated by eating an onion with the nose open and then eating it with the nose plugged.

When the chemoreceptor cells of taste buds are stimulated by the chemicals present in food or drink, they produce nerve impulses in associated sensory neurons. The nerve impulses travel along nerves (cranial nerve VII, IX, and X) and reach brainstem. From here, the impulses are transmitted to the taste-sensing areas of cerebral cortex. The cortex processes this information with other sensory information, such as smell and texture, to create the complete perception of taste.

Humans have four kinds of taste buds which respond to salty, sweet, sour, and bitter tastes. Taste buds for specific tastes are concentrated in specific regions of tongue: **sweet** at the tip, **sour** at the sides, **bitter** at the back, and **salty** over most of the tongue's surface. All the four regions overlap at certain places.

15.6.2- Receptors of Smell

The chemoreceptors located in the upper part of nasal cavities are responsible for the sense of smell (olfaction). These receptors are actually the ends of sensory neurons. These sensory neurons are present in a thin layer of tissue called nasal epithelium. They are packed tightly between two other types of cells: supporting cells,

which provide protection, and basal cells, which act as stem cells to replace old neurons.

The dendrites (the receiving ends) of these sensory neurons have tiny, hair-like structures called **cilia**. These cilia project outward into the moist mucus that lines the nasal cavity. When air enters the nasal cavities, the air-borne chemicals dissolve in the mucus. These dissolved chemicals bind to specific receptor sites on the cilia. This binding creates nerve impulse in sensory neurons, which is transmitted to the olfactory bulb through olfactory nerves (cranial nerve I). The olfactory bulb produces the perception of smell.

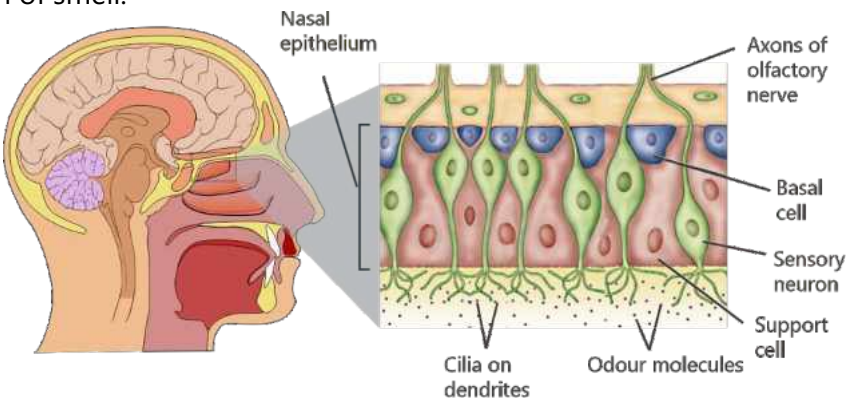


Figure 15.18: Olfactory receptors

15.6.3- Receptors of Pain & Touch

Different cutaneous receptors (receptors of the skin) respond to touch, pressure, heat, cold, and pain.

Nociceptors

Nociceptors detect pain. Pain is defined as a stimulus that causes or is about to cause tissue damage. Nociceptors consist of free nerve endings located throughout the body, especially near surfaces. Different nociceptors may respond to extremes in temperature, very intense pressure, or specific chemicals (including some that are released by injured cells).

Touch Receptors

The skin contains a variety of touch receptors. They are mechanoreceptors that respond to different physical stimuli.

For Information

Thermoreceptors: are naked dendrite endings of sensory neurons. They detect fall and rise in temperature. Cold receptors are located immediately below the epidermis, while warm receptors are located slightly deeper, in the dermis. Thermoreceptors are also found within the hypothalamus of the brain, where they detect the temperature of the blood and thus monitor the body's internal (core) temperature.

i- **Meissner's corpuscles** are endings of sensory neurons surrounded by a capsule. They are found in the upper layer of the skin (especially in lips, fingertips, palm, and soles). They detect light touch and vibration.

ii- **Hair follicle receptors** are endings of neurons and are located around the base of hair follicles. They detect the movement of hairs, which occur due to mechanical or thermal stimuli.

iii- **Merkel cells** are oval-shaped receptors in the deepest layer of epidermis. They detect light touch and are present in highly touch-sensitive areas like fingertips.

iv- **Pacinian corpuscles** are sensory neuron endings, surrounded by a capsule. They are located deeper in the skin. They detect deep pressure and vibration.

v- **Ruffini endings** are also sensory neuron endings enclosed in capsule. They are located in the dermis. They detect sustained pressure and stretching.

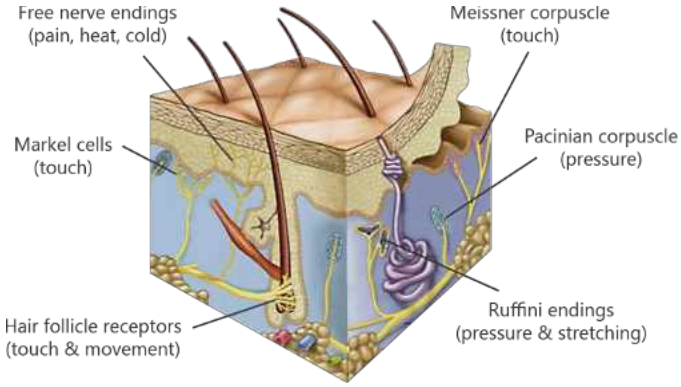


Figure 15.19: Touch and pain receptors

These touch receptors generate and transmit impulses via sensory neurons to the spinal cord and then to brain, where the signals are processed to create the sensations of touch.

15.7- Effects of Drugs on Nervous Coordination

A **drug** is defined any substance (other than food) that is used to prevent, diagnose, treat, or relieve symptoms of a disease or abnormal condition. Drugs can also affect the working of brain and cause changes in mood, awareness, thoughts, feelings, or behavior. A **psychoactive drug** is a type of drug that alters brain functions, resulting in changes in mood, perception, consciousness, and behaviour. Examples of psychoactive drugs include alcohol, caffeine, nicotine, and illicit drugs such as cocaine, heroin etc. A **narcotic drug** (or simply narcotic) is a type of opioid drug

For Information
All narcotics are psychoactive drugs, but all psychoactive drugs are not narcotics.

that is used for pain relief and has a high potential for addiction and abuse. Narcotics include drugs such as morphine, codeine, and oxycodone.

15.7.1- Addiction, Tolerance and Withdrawal Symptoms

The abuse of psychoactive drugs often leads to dependence. **Dependence** is a state in which a person relies on a drug physically or emotionally in order to function. Dependence often results in addiction. Drug **addiction** means the disorder of uncontrollable drug desire and use despite its negative effects.

As a person continues to use a drug, the body becomes adaptive and less sensitive to its effects. This condition is called drug **tolerance**. After drug tolerance, the person may require higher doses to achieve the same level of response. This leads to an increased addiction.

When addicts stop taking an addictive drug, they experience **withdrawal symptoms**. Withdrawal symptoms may include vomiting, headache, insomnia, breathing difficulties, depression, mental instability, and seizures.

Changes in Nervous System during Addiction and Tolerance

Addictive drugs affect the regions of brain which regulate pleasure and motivation. When these drugs reach brain, the presynaptic neurons release dopamine neurotransmitter, which produces feelings of pleasure and reward. The drug molecules block the dopamine reabsorption receptors in presynaptic neuron. So, large amount of dopamine accumulates in synaptic cleft. Brain adapts and reduces the number of dopamine receptors on postsynaptic neuron. This leads to tolerance i.e.; the user requires higher doses of drug to achieve the same pleasure or reward. When the drug is stopped abruptly, dopamine release becomes normal but due to less receptors, the postsynaptic neuron is less-stimulated and it leads to withdrawal symptoms.

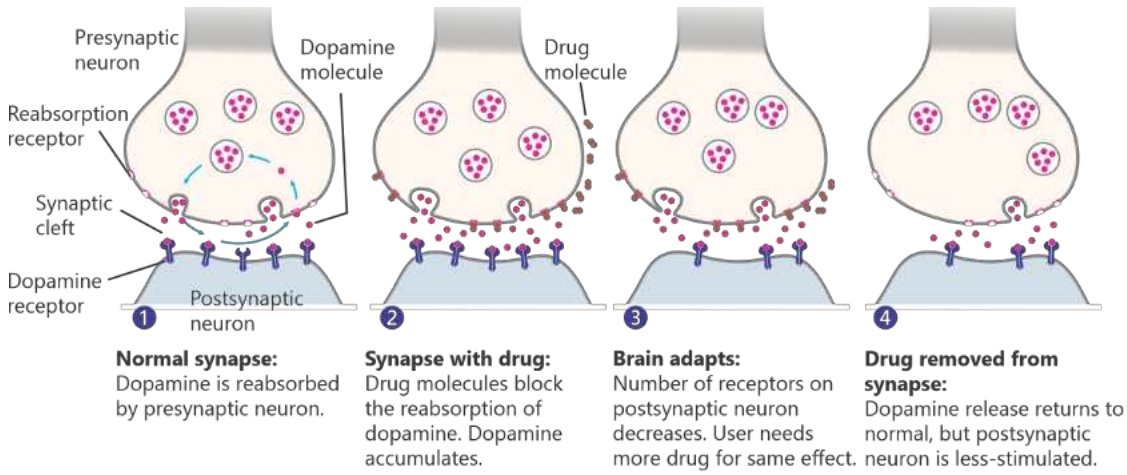


Figure 15.20: Neural changes during drug usage, addiction, tolerance, and withdrawal

Addiction and Tolerance of Caffeine

Caffeine is found in many common beverages, such as coffee, tea, and energy drinks. It can block the action of a neurotransmitter called adenosine, which promotes sleep and relaxation. As a result, caffeine increases alertness and energy levels.

Regular caffeine use can lead to tolerance. So, the user requires higher doses to achieve the same level of alertness. If caffeine use is stopped abruptly, withdrawal symptoms such as headaches and fatigue can occur.

Addiction and Tolerance of Nicotine

Nicotine is highly addictive stimulant present in tobacco products (cigarettes, cigars, and smokeless tobacco). It binds to nicotine receptors in brain. These receptors release dopamine and other neurotransmitters to produce pleasurable feelings.

Regular nicotine use leads to tolerance. So, the user requires higher doses to achieve the same level of pleasure. When nicotine use is stopped abruptly, the user experiences withdrawal symptoms, such as irritability, anxiety, and cravings. Long-term nicotine use is also associated with a variety of negative health effects, including an increased risk of cancer, heart disease, and respiratory problems.

15.7.2- Use and Abuse of Drugs

The term "use" refers to the appropriate or prescribed use of drugs for medical purposes. The term "abuse" generally refers to the inappropriate use of psychoactive drugs. The abuse of common psychoactive drugs is described next.

1- Heroin

It is a highly addictive opioid drug that is derived from the opium poppy plant. It is injected, smoked, or snorted. It produces false senses of euphoria (intense excitement and happiness) and relaxation. Its use quickly leads to dependence and addiction. It has a range of negative physical and mental health effects, including respiratory depression, infections, and mental health disorders.

2- Cannabis

Cannabis is a psychoactive drug that is made from dried flowering tops, leaves and stem of Indian hemp plant *Cannabis sativa*. It includes drugs like marijuana and hashish. It can be smoked, vaporized, or consumed in edibles. The use of cannabis produces sense of relaxation, euphoria, altered perception, and increased appetite. Its long-term use leads to negative effects, such as impaired cognitive function, respiratory problems, and mental health disorders.

3- Nicotine

Nicotine is a highly addictive drug found in tobacco products, including cigarettes, cigars, and smokeless tobacco. Its use leads to addiction and dependence,

as well as a range of negative physical and mental health effects, including respiratory problems, cancers, cardiovascular disease, and mental health disorders.

4- Alcohol

Alcohol is a psychoactive drug that is commonly consumed in various forms, such as beer, wine, and spirits. It produces feelings of relaxation and euphoria (intense happiness or excitement). Its negative effects include impaired cognitive and motor functions, liver diseases, cardiovascular diseases, and mental health disorders.

5- Inhalants

Inhalants, such as nail polish remover and glue, are psychoactive substances that are inhaled to produce senses of euphoria, relaxation, and altered perception. They can produce a range of negative health effects, such as respiratory problems, cognitive impairment, and neurological damage.

Withdrawal Symptoms of Drug Abuse

The withdrawal symptoms of drug abuse include anxiety, depression, fatigue, agitation, mood swings, shakiness or tremors, headache, sweating, nausea, loss of appetite, insomnia, rapid heart rate, high blood pressure, hallucinations (visual, auditory, or tactile), and seizures.

15.8- Disorders of Nervous System

15.8.1- Vascular Disorders

Vascular disorders of nervous system are the conditions in which the blood vessels of brain and spinal cord are affected. Examples include stroke and cerebral venous thrombosis.

Stroke

A stroke means damage in brain cells due to interruption in the blood supply to a part of brain. Such interruption may be due to a blockage or a rupture of a blood vessel of brain. **Causes** of stroke include high blood pressure, smoking, diabetes, high cholesterol, obesity, and family history of stroke. **Symptoms** of a stroke vary depending on the area of brain affected. Paralysis occurs on the side of body opposite the cerebral infarction (death of tissue). Other common symptoms are aphasia

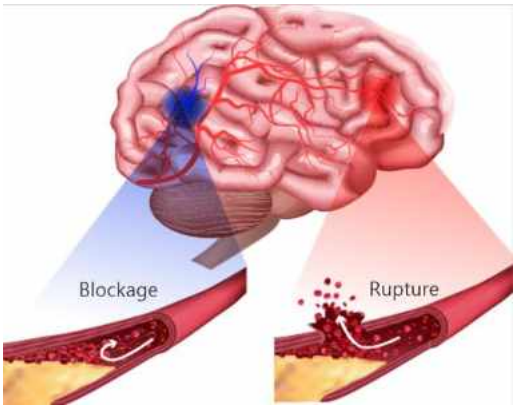


Figure 15.21: Main types of strokes

(inability to express through words); sudden trouble in seeing and walking, dizziness, loss of balance; and severe headache

For Information
Ischemic stroke occurs when the brain's blood vessels become narrowed or blocked
Haemorrhagic stroke occurs when a blood vessel in brain leaks or ruptures.
Transient ischemic attack (ministroke) occurs by a temporary decrease in blood supply to part of brain, which may last as little as five minutes.

Treatment should be given as soon as possible to minimize damage to brain. Treatment may include: medications (such as blood thinners, clot-dissolving drugs, or antihypertensive drugs); surgery to remove a blood clot or repair a ruptured blood vessel; and rehabilitation (to help patients recover physical and cognitive function)

15.8.2- Infectious Disorders

Infections of the central nervous system can be caused by almost any infectious agent, including viruses, bacteria, fungi, protozoa, and flatworms.

Meningitis

Meningitis is an inflammation of the meninges due to infection. **Causes:** Bacterial meningitis is the most severe and life-threatening. Viral meningitis is usually less severe. Fungal meningitis is rare and usually occurs in people with weak immune systems. Other causes include parasitic infections. **Symptoms** include stiffness in the neck, headache, and fever. In severe cases, meningitis may also cause paralysis, or coma. **Treatment:** Bacterial meningitis requires immediate treatment with antibiotics. Viral meningitis is usually self-limiting, although antiviral medications may be used. Fungal meningitis requires long-term treatment with antifungal medications. Vaccines are available for some of the bacterial meningitis, which can prevent infection.

15.8.3- Structural Disorders

Structural disorders of nervous system result from abnormalities or damage to the physical structure of nervous system. Examples include brain tumour, and spinal cord injuries.

Brain Tumour

A brain tumor is an abnormal growth or mass of cells in brain. Brain tumors can be benign (non-cancerous) or malignant (cancerous). Brain tumour can arise from glial cells or from the neurons themselves. **Causes:** The exact cause of brain tumors is unknown, but risk factors include exposure to radiation, family history, and certain genetic mutations. **Symptoms** depend on the location of tumour. These may include headaches, severe nerve pain, paralysis, seizures, and coma. **Treatment:** Common treatments include surgery, radiation therapy, and chemotherapy.

15.8.4- Functional Disorders

Functional disorders are the conditions that affect the normal functioning of the nervous system without any apparent structural or organic cause.

Headache

Headache is a common condition characterized by pain or discomfort in head or neck. Primary headaches are due to headache itself and not due to another cause e.g., migraine, tension headache. Secondary headaches are due to an underlying structural problem in head or neck such as bleeding in the brain, tumour, meningitis etc. **Treatments** include pain-killing (analgesic) medicines, and reducing stress, getting regular exercise, and improving sleep habits.

For Information
Migraine is a type of headache that is characterized by intense, throbbing pain on one side or in one region of head. Migraines are caused by changes in the brain and blood vessels, which lead to inflammation and pain.

15.8.5- Degenerative Disorders

These are the conditions in which there is progressive deterioration of nerve cells in brain, spinal cord, or peripheral nerves. Examples include **Parkinson's disease** (progressive death or weakening of the neurons in brain that produce dopamine leading to movement issues and cognitive changes), **Huntington's disease** (progressive damage to neurons in the brain, leading to movement disorders, cognitive decline, and psychiatric symptoms), multiple sclerosis (autoimmune disease of the CNS in which myelin sheath is damaged leading to vision loss, pain, fatigue, and impaired coordination), and Alzheimer's disease.

Alzheimer's Disease

Alzheimer's disease is a progressive degenerative brain disorder that affects memory, thinking, and behaviour. It leads to dementia in older adults. Its onset usually occurs in aged people, but it is not particularly associated with aging. **Causes:** It is due to a combination of genetic, environmental, and lifestyle factors. **Symptoms** typically begin with mild memory loss and confusion and progress to more severe cognitive impairment, including difficulty with language, problem-solving, and completing familiar tasks. **Treatment:** There is no cure for Alzheimer's disease, but there are medicines to slow its progression and to improve memory and cognitive function.

15.9- Diagnostic Tests for Nervous Disorders

A number of tests have been developed to diagnose the disorders of nervous disorders.

15.9.1- Electroencephalography (EEG)

EEG test measures the electrical activity of brain. Its basic principle is based on detecting the electrical nerve impulses being transmitted among the neurons.

Procedure: During EEG, metal electrodes are attached to the scalp. These electrodes are connected to an amplifier. The amplifier magnifies the electrical signals detected by the electrodes. The signals are then recorded and displayed as a series of **waveforms** on a computer screen or paper.

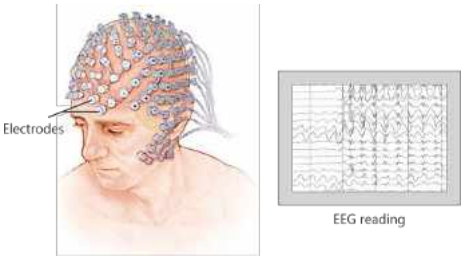


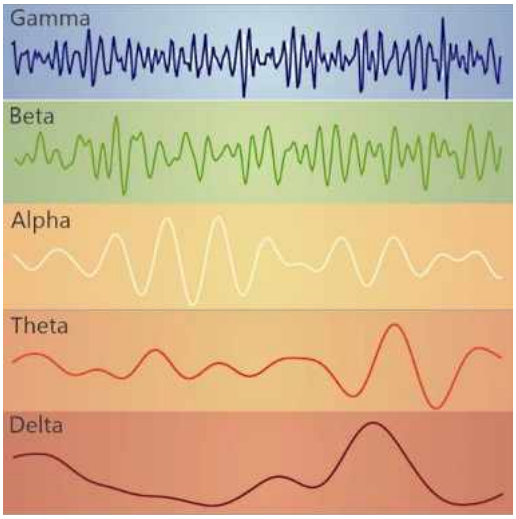
Figure 15.22: Electroencephalography

Uses: EEG is done to evaluate various neurological conditions, including seizures, epilepsy, and brain tumors. It is painless and does not require sedation or anaesthesia.

For Information

Each wave of EEG is associated with different states of brain activity.

- Gamma waves are associated with high perception, learning, problem-solving activities.
- Beta waves are associated with normal alert state and active thinking.
- Alpha waves show physically and mentally relaxed state.
- Theta waves show deep sleep (dreams and reduced consciousness).
- Delta waves show deepest sleep and dreaming.



15.9.2- Computed Tomography (CT)

CT scan is an imaging technique that uses X-rays and computer processing to generate detailed images of body's internal structures. The basic principle of CT is based on the fact that different tissues in body have varying degrees of X-ray

absorption. These differences can be used to create detailed images of internal structures.

Procedure: During a CT scan, the patient is on a motorized table that slides into a doughnut-shaped machine called a **gantry**. The gantry contains an X-ray tube that rotates around the patient. Multiple detectors on the opposite side of gantry record the absorption of X-rays through different tissues. This information is then used by a computer to construct a detailed, cross-sectional image of the internal structures.

Uses: CT scans are useful for imaging soft tissues, such as brain, lungs, and abdomen, as well as for detecting abnormalities in bone structure. CT scans are used to diagnose cancers, infections, and injuries.

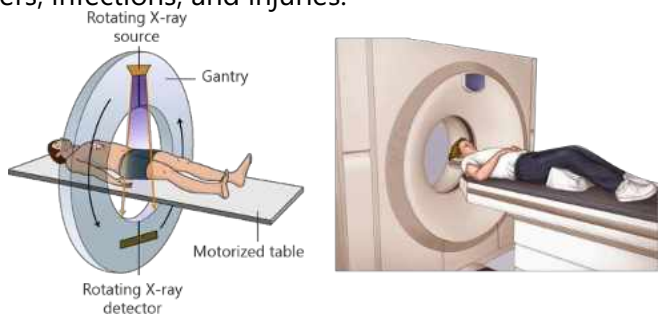


Figure 15.23: Computed tomography scan

15.9.3- Magnetic Resonance Imaging (MRI)

This imaging technique uses a strong magnetic field and radio waves to produce detailed images of body's internal structures. The basic principle of MRI is based on the fact that different tissues in the body have varying amounts of hydrogen atoms, which have magnetic properties.

Procedure: The patient is on a motorized table that slides into a large tube-like machine called a **scanner**. The scanner contains a powerful magnet that creates a strong magnetic field around the patient's body. Radio waves are then used to cause the hydrogen atoms in the body to produce a signal. These signals are then detected by a receiver. This information is used by a computer to construct a detailed, three-dimensional image of the internal structures.

Uses: MRI is used for imaging soft tissues, such as brain, spinal cord, and internal organs, as well as for detecting abnormalities in blood vessels and certain types of tumors.

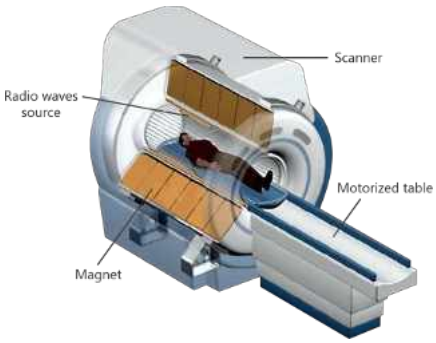


Figure 15.23: Magnetic Resonance Imaging

EXERCISE

SECTION 1: MULTIPLE CHOICE QUESTIONS

1. Neurotransmitter receptors are located on;
   (a) Nuclear membrane (b) Nodes of Ranvier
   (c) Postsynaptic membrane (d) Myelin sheath
2. Which neurotransmitter is released by all motor neurons at their synapses with skeletal muscles?
   (a) Acetylcholine (b) GABA (c) Norepinephrine (d) Dopamine
3. Which part of the nervous system prepares the body during emergency?
   (a) Central nervous system (b) Sympathetic nervous system
   (c) Parasympathetic nervous system (d) Sensory pathway
4. Which of these is closest to the brain tissue?
   (a) Acetylcholine (b) Arachnoid (c) Pia mater (d) Cranium
5. In meninges, the cerebrospinal fluid is present between;
   (a) Skull and dura mater (b) Dura mater and arachnoid
   (c) Arachnoid and pia mater (d) Pia mater and brain or spinal cord
6. Which is NOT a function of cerebrospinal fluid?
   (a) Cushions brain (b) Produces hormones
   (c) Transport nutrients to brain (d) Protects the spinal cord
7. During saltatory conduction, the nerve impulse jumps from one _____ to another.
   (a) Axon (b) Synapse (c) Myelin sheath (d) None of Ranvier
8. In a resting neuron, which of these ions has the highest permeability?
   (a) Chloride (b) Sodium (c) Potassium (d) Calcium
9. If nerve impulses in a neuron 'a' are travelling faster than the impulses in neuron 'b', what might be the reason?
   (a) Hyperpolarization in neuron 'a'
   (b) Neuron 'a' is myelinated
   (c) There is a high concentration of sodium ions inside neuron 'a'
   (d) Neuron 'a' is thicker than neuron 'b'
10. Dorsal root of spinal nerve contains?
    (a) Sensory neurons (b) Inter-neurons
    (c) Motor neurons (d) Sensory and motor neurons
11. Which ions cause the presynaptic vesicles to fuse with the presynaptic membrane and release a neurotransmitter into the synaptic area?

(a) Sodium (b) Potassium (c) Calcium (d) Iodine

12. The most common cause of dementia is;
    (a) Alzheimer's disease (b) Brain tumour
    (c) Stroke (d) Meningitis
13. Nociceptors are specialized receptors that detect:
    (a) Light touch (b) Deep pressure (c) Pain (d) Taste
14. Which receptor detects deep pressure and vibration?
    (a) Meissner's corpuscles (b) Pacinian corpuscles
    (c) Merkel cells (d) Ruffini endings
15. Which of the following is a narcotic drug used for pain relief?
    (a) Caffeine (b) Morphine (c) Alcohol (d) Nicotine
16. Withdrawal symptoms of drug abuse include:
    (a) Improved concentration (b) Headache, nausea, and anxiety
    (c) Increased euphoria (d) Faster drug metabolism
17. Caffeine increases alertness by:
    (a) Increasing dopamine receptors (b) Blocking adenosine receptors
    (c) Stimulating nociceptors (d) Enhancing serotonin degradation

SECTION 2: SHORT QUESTIONS

1. Enlist the basic elements of nervous coordination.
2. Define receptors and effectors with examples.
3. State the path of a message transmitted through the nervous system.
4. Define nerve impulse.
5. Name the factors responsible for the resting membrane potential of neuron.
6. What are the main components of the nervous system?
7. Define narcotic drugs and give examples.
8. Briefly describe the effects of drug addiction and tolerance on the functioning of nervous system.
9. State the causes, symptoms and treatment of stroke and meningitis.
10. State the causes, symptoms and treatment of brain tumor.
11. State the causes, symptoms and treatment of Alzheimer disease.
12. Differentiate between;
    - Axon and dendrite
    - Sensory neuron, inter-neuron, and motor neurons
    - Myelinated and nonmyelinated neurons
    - White matter and grey matter
    - Cranial and spinal nerves

- Resting membrane potential and active membrane potential
- Polarization and depolarization
- Repolarization and hyperpolarization
- Presynaptic and postsynaptic neuron
- Synaptic knob and synaptic vesicles
- Somatic and autonomic nerves system
- Drug addiction and drug tolerance

SECTION 3: LONG QUESTIONS

1. Explain the structure of a neuron.
2. Explain the function of the three types of neurons with the help of a reflex arc.
3. Describe the generation and transmission of nerve impulse.
4. Describe polarization, depolarization and hyperpolarisation of neuron membrane.
5. Describe the saltatory conduction of nerve impulse.
6. Describe the structure of synapse.
7. Explain synaptic transmission of nerve impulse.
8. Classify neurotransmitters as inhibitory and excitatory and list some common examples.
9. Explain briefly the functions of major parts of brain.
10. Describe the structure of human brain and compare it with that of spinal cord.
11. Explain the types and functions of autonomic nervous system.
12. Write a detailed note on the receptors for smell.
13. Explain the structure of the receptors for taste.
14. Describe the receptors for touch and pain.
15. Describe the use and abuse of narcotic drugs with examples.
16. What is meant by drug addiction and drug tolerance? Explain with reference to caffeine and nicotine.
17. Explain the classification of nervous disorders with examples.
18. Explain the principles and use of EEG, CT scan and MRI.

INQUISITIVE QUESTIONS

1. Predict from every day experience what various kinds of receptor can be found in human body.
2. Conceptualize the electrical activity of brain, which can be recorded using magnets and tomography.
3. Justify the way nervous system helps to coordinate complex and intricate movements of hand to play a piano, or write alphabets.
4. Justify that the development of a modern computer is in fact a product of the understanding of the way nervous coordination occurs in complex organisms like humans.

Chapter 16 HUMAN ENDOCRINE SYSTEM

STUDENTS'LEARNING OUTCOMES

After studying this chapter, the students will be able to:

- State the role of hormones as chemical messengers.
- Describe the chemical nature of hormones and correlate it with important hormones.
- Locate the endocrine glands in human body name the hormones they release and their functions; (pituitary, thyroid, parathyroid, pancreas, adrenal, gonads).
- Relate the problems associated with their imbalance of these hormones.
- Explain the neurosecretory role of hypothalamus.
- Outline the concept of feedback mechanism of hormones and describe positive feedback with reference to Oxytocin and negative feedback with reference to Insulin and Glucagon.

While our nervous system carries out high-speed nervous coordination, the endocrine system controls long-term processes like growth, metabolism, and reproduction. It performs its job by sending chemical instructions, in the form of hormones, to the target body parts. It is called chemical coordination. In this chapter we will study the hormones of endocrine glands, and their functions.

16.1- Hormones – The Chemical Messengers

A hormone is a regulatory chemical that is secreted into the blood or tissue fluid by an endocrine gland. The blood carries the hormone to every cell in body, but it shows its effects only in its specific target cells. When the target cells receive a hormone, they respond in specific way. That's why, hormones are referred to as **chemical messengers** or signalling molecules.

For Information

Glands are the tissues that produce and release secretions. There are two types of glands in body. Exocrine glands secrete their secretion in ducts attached with them. While, endocrine glands are ductless. They secrete their secretions called hormones directly in blood or tissue fluid.

16.1.1- Chemical Nature of Hormones

We can group hormones into three main chemical categories.

1. Peptide and Protein Hormones

These are made of chains of amino acids. They are water-soluble, meaning they dissolve easily in blood. However, they cannot pass through cell membranes. So, they bind to receptors on the cell surface. Examples include;

Hormone	Source (Gland)	Main Function
Insulin	Pancreas	Lowers blood glucose levels.
Glucagon	Pancreas	Raises blood glucose levels.
Growth Hormone	Anterior Pituitary	Stimulates growth and cell reproduction.
Oxytocin	Hypothalamus (released by Posterior Pituitary)	Stimulates muscle contractions during childbirth.

2. Glycoprotein Hormones

Glycoproteins are "conjugated proteins". They consist of a protein combined with a carbohydrate (sugar) group. They are large, complex, and polar molecules. So, they also cannot pass through the cell membrane. Examples include;

Hormone	Source (Gland)	Main Function
Thyroid Stimulating Hormone	Anterior Pituitary	Stimulates the thyroid gland to produce thyroxine.
Follicle Stimulating Hormone	Anterior Pituitary	Stimulates sperm production or egg development.
Luteinizing Hormone	Anterior Pituitary	Triggers ovulation in females and secretion of testosterone in males.

3. Amino Acid Derivatives Hormones

These are small molecules derived from a single amino acid (usually Tyrosine or Tryptophan). Their ability to pass through cell membranes depends on whether they behave more like a protein or more like a lipid. Epinephrine and norepinephrine cannot cross membranes while thyroxine and tri-iodothyronine can cross. Examples include;

Tidbit

In 1902, two British scientists, Bayliss and Starling, discovered hormone during their research on digestive system. They found that when acidic gastric juice comes in small intestine, a substance (which they called “secretin”) was released into blood. Secretin reaches the pancreas and stimulates it to secrete pancreatic juice.

Hormone	Source (Gland)	Main Function
Epinephrine (Adrenaline)	Adrenal Medulla	Immediate "fight or flight" response.
Norepinephrine (noradrenaline)	Adrenal Medulla	Increases blood pressure and alertness.
Thyroxine (T4)	Thyroid Gland	Controls the speed of metabolism.
Triiodothyronine (T_3)	Thyroid Gland	Regulates body temperature and heart rate.

4. Steroid Hormones

These hormones are derived from cholesterol (a type of lipid). Unlike proteins, steroids are lipid-soluble (fat-soluble). This means they can move through the cell membrane to give instructions directly to the nucleus. Examples include;

Hormone	Source (Gland)	Main Function
Cortisol	Adrenal Cortex	Regulates metabolism and helps the body respond to stress.
Aldosterone	Adrenal Cortex	Helps the kidneys manage salt and water balance.
Testosterone	Testes	Regulates male reproductive tissues and traits.
Estrogen / Progesterone	Ovaries	Regulate the female reproductive system and cycle.

Path of the chemical message carried by hormones

The endocrine glands secrete their hormones on receiving particular stimuli. They release hormones in blood, which carries the hormone to all cells. Each hormone affects only its target cells. Target cells have receptors. Each receptor binds to a specific hormone. When a hormone binds to a receptor, the binding triggers events that lead to changes within the cell. Receptors can be found on the cell membrane, in the cytoplasm, or in the nucleus of a cell.

Tidbit
Hormones and neurotransmitters are both chemical messengers. However, hormones are part of endocrine system, while neurotransmitters are part of nervous system. Hormones are often slower acting and have longer effects than neurotransmitters.

16.2- Endocrine System of Man

The endocrine system of human beings consists of glands and organs that produce and secrete hormones for regulating various body functions. The locations of major endocrine glands have been shown below.

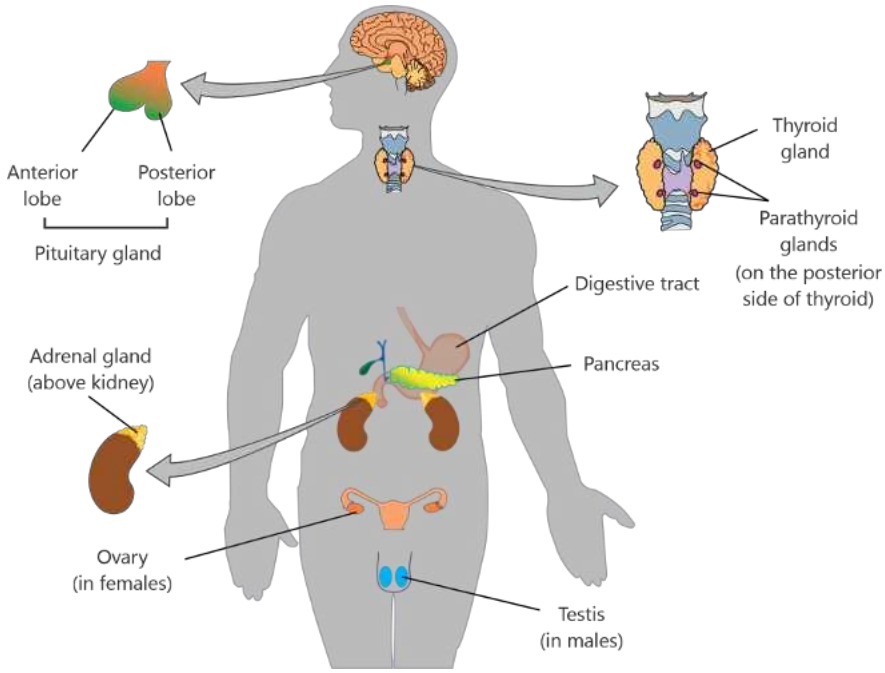


Figure 16.1: Locations of major endocrine glands in human body

16.2.1- Neurosecretory Role of Hypothalamus

Many endocrine glands of the body are controlled by the master gland i.e., pituitary gland. However, the master gland is itself controlled by a part of the brain i.e., hypothalamus. Hypothalamus contains special neurosecretory cells, which not only conduct nerve impulses but also secrete hormones.

Control of Posterior Pituitary

Two hormones i.e., oxytocin and antidiuretic hormone (ADH) are made by neurosecretory cells of hypothalamus. The axons of neurosecretory cells transport oxytocin and ADH to the posterior pituitary, where they are stored for eventual release into blood.

Control of Anterior Pituitary

Blood vessels connect hypothalamus with the anterior pituitary. Neurosecretory cells of hypothalamus secrete hormones that travel to the anterior pituitary through these blood vessels. These hormones are of two kinds

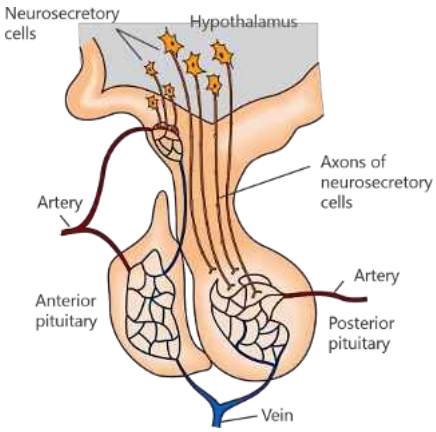


Figure 16.2: Hypothalamus and pituitary

- **Releasing hormones** stimulate the anterior pituitary to make and secrete hormones.
- **Inhibiting hormones** inhibit production and secretion of anterior pituitary hormones.

Table: Hypothalamic hormones and their effect on pituitary gland

Hormones of hypothalamus	Effect on Anterior pituitary
Growth Hormone Releasing-Hormone	Secretion of growth hormone
Somatostatin	Inhibition of growth hormone
Thyrotropin releasing hormone (TRH)	Secretion of thyroid stimulating hormone (TSH)
Adreno-corticotropin releasing hormone	Secretion of adrenocorticotrophic hormone
Prolactin inhibiting factor – which is actually dopamine	Inhibits secretion of prolactin
Gonadotropin releasing hormone	Secretion of FSH and LH

16.2.2- Pituitary Gland

Pituitary gland is attached to hypothalamus by a stalk called **infundibulum**. Pituitary gland is divided into three lobes called the anterior pituitary, posterior pituitary and intermediate / median pituitary.

Anterior Pituitary

Anterior pituitary secretes many hormones, many of which regulate the activity of other endocrine glands.

1. **Growth Hormone** (GH, or somatotropin) stimulates the growth of muscles, bones (indirectly), and other tissues. It is also essential for proper metabolic regulation.
2. **Adrenocorticotropic Hormone** (ACTH, or corticotropin) stimulates the adrenal cortex for the secretion of corticosteroids (cortisone and aldosterone), which regulate glucose and salt homeostasis.
3. **Thyroid-stimulating Hormone** (TSH, or thyrotropin) stimulates the thyroid gland to produce thyroxine, which in turn stimulates oxidative respiration.
4. **Luteinizing Hormone** (LH) In females, it stimulates ovaries to produce progesterone and estrogen hormones. It

Tidbit

Growth Hormone Treatment

GH treatment is given to children have short stature due to GH deficiency. It improves their growth rate.

However, every dwarf person does not have GH deficiency. So, GH therapy may not be appropriate for everyone.

Some people use GH because they believe it will build muscle or slow aging. However, doctors do not support the use of GH for these purposes.

also initiates ovulation. In males, it stimulates testes for testosterone production.

5. **Follicle-stimulating Hormone** (FSH) stimulates the development of ovarian follicles in females. In males, it stimulated the development of sperms. FSH and LH are both referred to as gonadotropins.
6. **Prolactin** stimulates the mammary glands to produce milk. It is produced in both male and female but functions only in female.

Posterior Pituitary

The posterior pituitary contains axons that originate in hypothalamus and extend to posterior pituitary. It does not synthesize any hormone but only stores and releases the hormones made by hypothalamus. The following are the hormones released by posterior pituitary.

1. **Antidiuretic Hormone** (ADH or vasopressin) stimulates water retention by the kidneys.
2. **Oxytocin** stimulates the milk-ejection by stimulating the contraction of smooth muscles of mammary glands. It also stimulates uterine contractions in women during childbirth.

For Information
During embryonic development, posterior pituitary is formed from an outgrowth of neural tissue. That's why, hypothalamus of brain and posterior pituitary remain interconnected by a tract of axons.

For Information
Oxytocin secretion continues after childbirth in breast-feeding mothers. That's why, the uterus of a nursing mother returns to its normal size after pregnancy more quickly than the mothers who do not breast-feed.

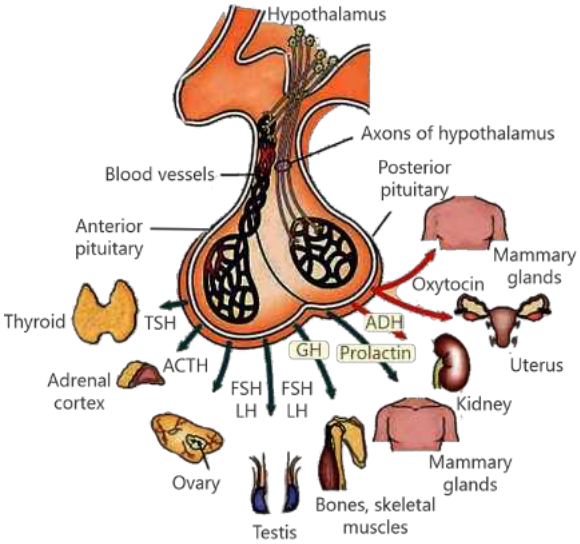


Figure 16.3: Hormones of anterior & posterior pituitary and their target sites

Intermediate Pituitary

In humans, intermediate pituitary is a thin layer of cells between the anterior and posterior pituitary. It produces **melanocyte stimulating hormone** (MSH). MSH stimulates the production and release of melanin by melanocytes in skin and hair.

Imbalance of Pituitary Hormones

1. If there is an over-secretion of growth hormone (GH) in a growing child, it causes **gigantism** i.e., height that is well above average, enlarged hands, feet, and facial features. Over-secretion of GH in adult life causes **acromegaly** in which bones are no longer capable of increasing in length but grow in thickness. Acromegaly is characterised by enlarged hands, feet, skull, nose and jawbone.
2. Deficiency of GH results in **dwarfism** in which development is much slower and individual has short stature. However, the body parts stay in proportion and brain development and IQ are unaffected.
3. Over secretion of TSH causes **hyperthyroidism** i.e., excess of thyroxine and its under secretion causes **hypothyroidism** i.e., lack of thyroxine.
4. Under secretion of ADH causes **diabetes insipidus**. It is characterized by excessive production of dilute urine and frequent thirst. Over secretion of ADH may lead to kidney problems.
5. Over secretion of oxytocin causes rupturing of uterine wall while under secretion of oxytocin inhibits normal labour process.

16.2.3- Thyroid Gland

It is a butterfly-shaped gland located on either side of the trachea in the front of neck. It is composed of two connected lobes. It secretes following hormones;

Tidbit
Thyroid gland is often called the body's "Master Controller of Metabolism" because its hormones dictate how fast or slow every cell in body works.

1. **Tetra-iodothyronine (T_4) or thyroxine and tri-iodothyronine (T_3):** Thyroid produces much more T_4, which eventually converts into T_3, which is the more "active" and powerful. These hormones increase heart rate and breathing rate. They stimulate cellular respiration and set the body's basal metabolic rate. They also enhance glucose catabolism and synthesis of cholesterol in liver. In children, they promote growth of the skeleton and the development of brain.
2. **Calcitonin** hormone: It plays a role in maintaining proper levels of calcium in blood. When the blood calcium concentration rises too high, calcitonin stimulates the absorption of calcium into bones, thus lowering its level in blood.

For Information
Graves' disease is an autoimmune disease that causes hyperthyroidism. It also often results in an enlarged thyroid. Its symptoms include irritability, muscle weakness, sleeping problems, fast heartbeat, poor tolerance of heat, diarrhoea, weight loss, thickening of skin on the shins. Exophthalmia (protrusion of the eyeballs) may also result from Graves' disease.

Imbalance of Thyroid Hormones

1. The deficiency of thyroid hormones (**hypothyroidism)** causes growth retardation, lethargy, weight gain, constipation, high cholesterol, and low heart rate and body temperature.
2. If the hypothyroidism occurs during foetal and childhood development, it causes **cretinism** i.e., a condition of stunted growth and mental retardation.
3. Severe hypothyroidism result in is a rare, life-threatening complication known as **myxoedema**. It is characterized build-up of mucopolysaccharides in the connective tissues, which causes swelling and thickening of the skin and tissues.
4. If hypothyroidism is caused by iodine deficiency, it results in **goitre** i.e., swelling of thyroid gland.
5. Excessive secretion of thyroid hormones **(hyperthyroidism)** causes weight loss; and high blood pressure, heart rate, and body temperature.

16.2.4- Parathyroid Glands

These are four small glands attached to the posterior surface of thyroid gland. Parathyroid glands secrete hormone **parathormone**. When blood calcium level falls, parathormone is released. It stimulates the osteoclasts (bone cells) to dissolve the calcium crystals of bone matrix and release calcium into the blood. It also stimulates the kidneys to reabsorb calcium from the urine. It causes the activation of vitamin D, needed for the absorption of calcium from food in the intestine.

Imbalance of Parathyroid Hormone

Over secretion of parathormone (which may be due to parathyroid gland tumour) results in excessive release of calcium from bones. It leads to bones deformation and soft bones. Blood calcium level elevates (**hypercalcemia**) which depresses nervous system and causes weakness of muscles. Under secretion of parathormone causes **hypocalcaemia**. This increases the excitability of neurons. Also, it can lead to tetany in which muscles remain in contracted state.

16.2.5- Pancreas

Pancreas mostly contains exocrine cells, which produce and secrete pancreatic juice in pancreatic duct. But pancreas also has specialized cells, which function as an endocrine gland. These cells are distributed in the form of patches in the pancreas. These patches are called Islets of Langerhans. Each islet is composed of two types of cells. **Beta (β) cells** are larger in number and they secrete insulin hormone. **Alpha (α) cells** secrete glucagon hormone.

Insulin is one of the most important hormones of human body. It reduces the blood sugar (glucose) level. It is secreted when the level of blood glucose rises. It increases the rate of glucose uptake by body cells (e.g., skeletal muscles and fat cells). It also promotes **glycogenesis** (conversion of glucose to glycogen) in liver, increases the use of glucose in cellular respiration, promotes the conversion of excess glucose to fats and inhibits **gluconeogenesis** (glucose synthesis).

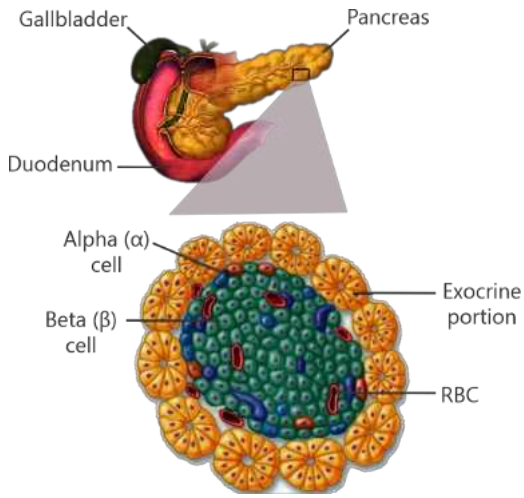


Figure 16.4: Islets of Langerhans (surrounded by exocrine portions)

Glucagon raises the blood glucose level. It is secreted when blood glucose level is below normal. It mainly promotes the hydrolysis of stored glycogen in liver. As a result, glucose is released into blood and can be taken up by cells and used for energy.

Imbalance of Pancreas Hormones

1. The under secretion of insulin leads to **diabetes mellitus (hyper-glycaemia)** in which cells are unable to obtain glucose from blood. It results in above normal blood glucose level. This excess glucose in blood inhibits water reabsorption by the kidneys. So large amounts of urine are produced. It leads to dehydration and kidney damage.
2. The over secretion of insulin results below normal blood glucose level i.e., **hypoglycaemia**. Symptoms of hypoglycaemia include lethargy, dizziness, nervousness, and in extreme cases, unconsciousness.

For Information

In **Type I diabetes** (insulin-dependent diabetes mellitus), the patients lack insulin-secreting β cells. It is generally is treated with daily injections of insulin (because insulin is a peptide hormone, it would be digested if taken orally and must be injected into blood). This injectable insulin is produced by genetically engineered bacteria.

The patients of **Type II diabetes** (non-insulin dependent diabetes mellitus) have normal levels of insulin in their blood, but their cells have a reduced sensitivity to insulin. These patients do not require insulin. They control their diabetes through other medicines, diet and exercise.

16.2.6- Adrenal Glands

One adrenal gland is located just above each kidney. Each gland is composed of an inner portion, the adrenal medulla, and an outer layer, the adrenal cortex.

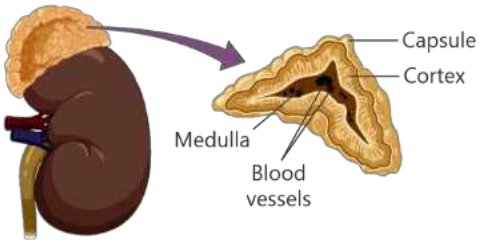


Figure 16.5: Adrenal gland

Adrenal Medulla

The adrenal medulla produces two hormones i.e., **epinephrine** and **norepinephrine** (also known as adrenaline and noradrenaline, respectively). When a person is stressed, the medulla secretes epinephrine and norepinephrine. The hormones prepare the body for "fight or flight". In this response, there is increased heart rate, increased blood pressure, dilation of the bronchioles, elevation in blood glucose, and reduced blood flow to skin and digestive organs.

For Information
Secretion of hormones from adrenal medulla is controlled by nervous system, whereas secretion of hormones from adrenal cortex is controlled by the hormones of anterior pituitary.

Adrenal Cortex

The hormones from adrenal cortex are called **corticosteroids**. These include:

1. **Glucocorticoids** e.g., cortisol and related steroids maintain glucose homeostasis. They stimulate the breakdown of muscle protein into amino acids, which are carried by blood to liver. They also stimulate liver for gluconeogenesis (conversion of amino acids into glucose).
2. **Mineralocorticoids** e.g., aldosterone helps to regulate mineral balance in blood. It stimulates the kidneys to reabsorb sodium from urine. In this way, water is also reabsorbed from urine. Thus, normal blood volume and pressure is maintained. Aldosterone also stimulates the kidneys to secrete the excess of potassium into urine.

Imbalance of Adrenal Gland Hormones

1. Over secretion of hormones of adrenal medulla may cause hypertension and aggressive behaviour during routine life. Under secretion causes failure to combat with emergency situation.
2. Over secretion of cortisol from adrenal cortex causes **Cushing disease**. It occurs when pituitary gland produces too much ACTH, which causes the adrenal glands to produce cortisol in high levels. In Cushing disease there is excessive protein

breakdown. It results in high blood sugar, high blood pressure, thin bones, muscle loss, thin and fragile skin, depression, and too much facial hair (in women).

3. Under secretion of cortisol leads to **Addison's disease**. It is characterized by general metabolic disturbance, weakness of muscles and loss of salts. The symptoms usually happen slowly.

16.2.7- Gonads

Gonads (ovaries in females and testes in males) are gamete-producing organs. They also produce sex hormones under the influence of LH and FSH of anterior pituitary.

Ovaries

Ovaries secrete female sex hormones i.e., estrogen and progesterone.

1. **Estrogen** is secreted by Graafian follicle in response to FSH. Its functions include development of secondary sexual characteristics (e.g., breast development, widening of the pelvis, fat distribution in hips and thighs etc.), regulation of the menstrual cycle, repair of the uterine lining after menstruation, ovulation (release of an egg from ovary), maintenance of bone density, lowering "bad" cholesterol (LDL), maintenance of healthy blood pressure, regulation of mood and memory.

For Information

Puberty is the adolescent stage during which the sex organs mature and secondary sex characteristics appear. During puberty in males, sperm production begins, the voice deepens, the chest broadens, and hair grows on the body and face. In females, the menstrual cycle begins, the breasts grow, and the hips widen.

2. **Progesterone** is produced by corpus luteum in response to LH during menstrual cycle. It is also produced and released from placenta during pregnancy. It inhibits further FSH secretion from pituitary, thus preventing more follicles from ripening. It causes further thickening and vascularisation of the uterus wall for maintaining pregnancy. Progesterone suppresses ovulation.

For Information

Small amounts of estrogen and testosterone are also released by adrenal glands.

A very small amount of testosterone is produced in females.

Testes

LH stimulates the testes to secrete a group of sex hormones called **androgens**. **Testosterone** is an androgen which is secreted by the interstitial cells among seminiferous tubules. It initiates the maturation of male reproductive organs and

appearance of secondary sex characteristics and sex drive. In addition, testosterone is necessary for normal sperm production.

Imbalance of Hormones of Gonads

1. Due to estrogen deficiency in the young females, they fail to mature sexually. Deficiency of estrogen in adults leads to sterility. Its over-secretion may lead to the development of **fibroids** (abnormal growth) in uterus and **polycystic ovaries**.
2. Under secretion of progesterone during menstrual cycle, decreases the chance of pregnancy and may cause early menstruation. Under secretion during pregnancy may leads to the **miscarriage**.
3. Under secretion of testosterone in males causes the development of feminine characteristics and male sterility.

For Information

Artificially Synthesized Steroids

Artificially synthesized steroids, also known as anabolic steroids, have been widely used by athletes to enhance performance in sports. These steroids are synthetic versions of the testosterone. They increase muscle mass, strength, and endurance. However, the use of anabolic steroids in sports is illegal and banned. It can lead to long-term health problems including:

- Anabolic steroids can cause damage to liver, leading to hepatitis and liver tumors.
- They can increase the risk of heart disease, stroke, and blood clots.
- Prolonged use of can disrupt the body's natural hormone balance, leading to infertility, impotence, and breast development in men.
- Their use can also cause mental health issues such as depression, anxiety, and aggressive behaviour.

16.2.8. Other Endocrine Glands/Tissues

There are several other glands in the endocrine system. There are also specialized endocrine cells in brain, stomach, small intestine, liver, and other organs.

1. **Thymus gland** is located beneath the sternum and between the lungs. It secretes **thymosin** hormone that stimulates maturation of T cells, which help defend the body from pathogens.
2. **Pineal gland** is located near the base of brain. It secretes **melatonin** hormone. It regulates sleep patterns.

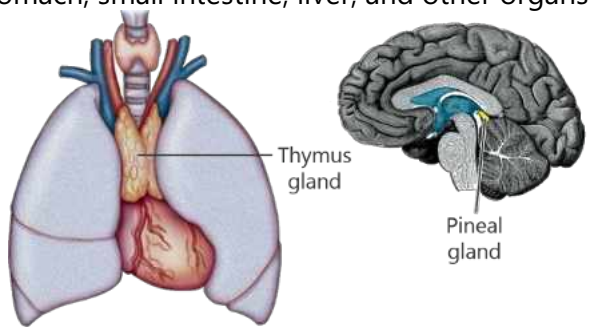


Figure 16.6: Locations of thymus and pineal glands

3. Endocrine cells within the walls of some **digestive organs** also secrete hormones that control digestive processes. For example, when food is eaten, endocrine cells in the stomach walls secrete **gastrin** hormone that stimulates the gastric glands to release gastric juice. Endocrine cells of the small intestine wall release **secretin** hormone that stimulates the pancreas to release pancreatic juice. The small intestine wall also secretes **cholecystokinin** hormone which stimulates gallbladder to release bile.
4. **Kidneys** produce **renin** hormone that controls the production of aldosterone in adrenal glands. Kidneys also produce **erythropoietin** hormone that stimulates bone marrow for the production of RBCs. Another hormone of kidneys, **calcitriol** increases blood calcium level by absorption of calcium from intestines.
5. **Brain** produces hormones called **endorphins**. Endorphins bind to pain receptors and so block sensation of pain.
6. **Prostaglandins** are hormone-like substances that are produced by almost all cells of the body. They are powerful, locally-acting vasodilators and are involved in inflammation.

16.3- Feedback Mechanism in Endocrine System

You have studied the concept of feedback mechanism in homeostasis. Feedback mechanism is the phenomenon in which a controlling mechanism is itself controlled by the product of the process it is controlling. Endocrine system also has feedback mechanisms, which control it. There are two types of feedback mechanism.

1- Positive Feedback

In positive feedback, the product or response made by the controlling system speeds up action of controlling system. An example of positive feedback is release of oxytocin hormone by posterior pituitary. During childbirth when uterus begins to contract, the sensory neurons send signals to hypothalamus. Hypothalamus triggers the release of oxytocin from posterior pituitary.

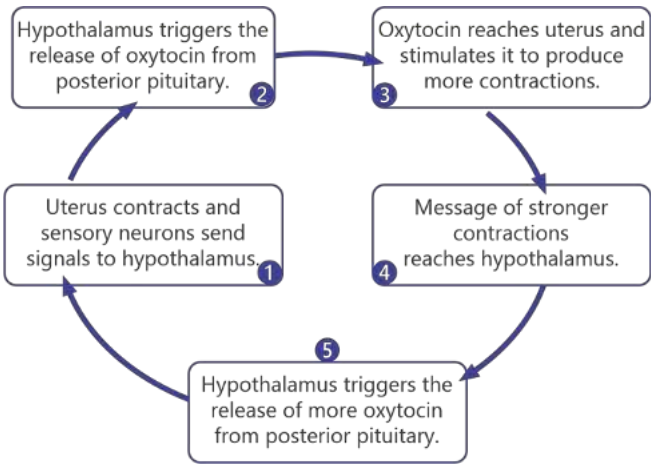


Figure 16.7: Positive feedback of oxytocin

Oxytocin stimulates uterus to produce more and stronger contractions. When hypothalamus receives message of stronger contractions, it triggers the release of more oxytocin. This series of positive feedback continues until the birth of baby.

2- Negative Feedback

In negative feedback, the end product stops the initial step of the process. Negative feedback is common in endocrine system. For example, when blood glucose goes above normal, the beta cells of the islets of Langerhans sense release insulin. Insulin lowers blood glucose by doing different actions. Due to these actions, blood glucose level falls to normal. This result i.e., normal blood glucose level stimulates the beta cells to stop releasing insulin. Similarly, the release of glucagon is also controlled by negative feedback. When blood glucose level goes low, the alpha cells of the islets of Langerhans secrete glucagon. This hormone takes actions to raise blood glucose. Due to these actions, blood glucose level rises to normal. This result i.e., normal blood glucose level stimulates the alpha cells to stop releasing glucagon.

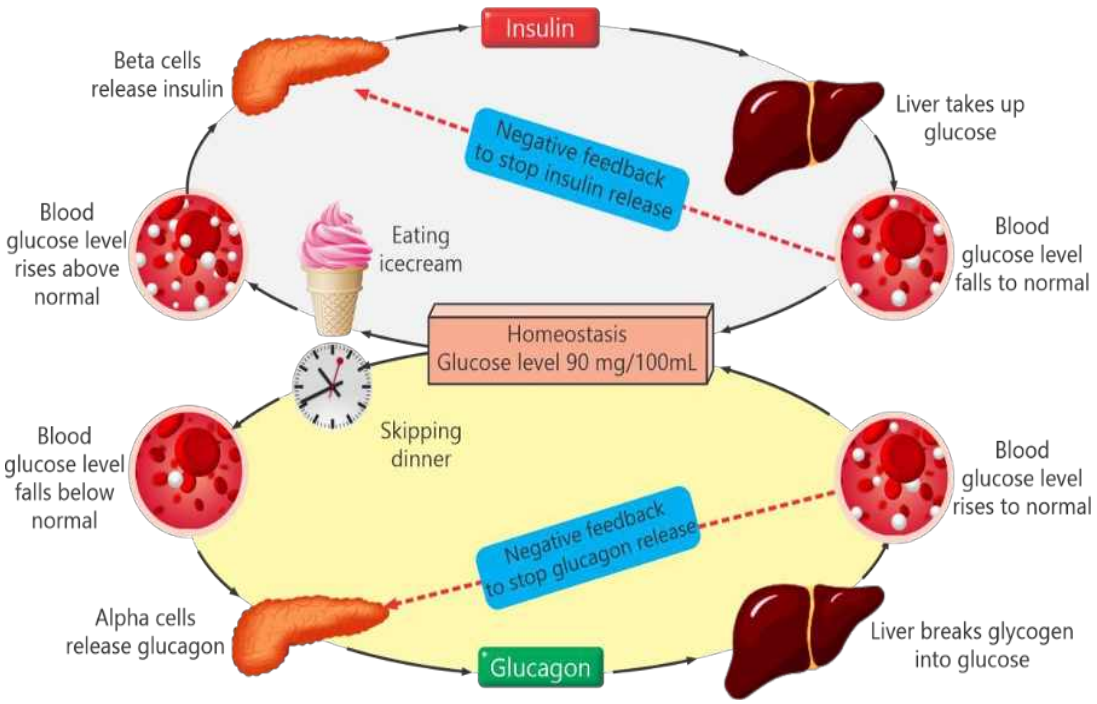


Figure 16.8: Negative feedback of insulin and glucagon

EXERCISE

SECTION 1: MULTIPLE CHOICE QUESTIONS

1. Which of the following pair of hormones has antagonistic (opposing) effects?
   (a) Parathyroid hormone and calcitonin (b) Growth hormone and epinephrine
   (c) Estrogen and progesterone (d) Cortisol and aldosterone
2. Which gland produces hormones that control other endocrine glands?
   (a) Anterior pituitary (b) Posterior pituitary
   (c) Thyroid (d) Intermediate pituitary
3. Which of the following is NOT released by anterior pituitary?
   (a) Prolactin (b) ADH (c) FSH (d) TSH
4. Antidiuretic and oxytocin hormones are produced by
   (a) Anterior pituitary (b) Posterior pituitary
   (c) Intermediate pituitary (d) Hypothalamus
5. Which of the following hormones binds to the receptors located inside the cell?
   (a) FSH (b) Testosterone (c) Insulin (d) Growth hormone
6. Which of the following is a function of a hormone released by the posterior pituitary?
   (a) Maturation of egg and sperm (b) Decrease in calcium levels
   (c) Water retention (d) Increase in thyroid hormone level
7. Which of the following hormones would be expected to increase if you were studying all day for a test and skipped breakfast and lunch?
   (a) Glucagon (b) Insulin (c) Growth hormone (d) Calcitonin
8. Which of the following is both an endocrine and exocrine gland?
   (a) Anterior pituitary (b) Pancreas
   (c) Thyroid (d) Adrenal medulla
9. Which hormone increases basal metabolic rate in the body?
   (a) Thyroxine (b) Parathormone (c) Glucagon (d) Secretin
10. Which of the following is NOT a site of action of insulin?
   (a) Smooth muscles (b) Skeletal muscles
   (c) Adipose tissue (d) Liver

SECTION 2: SHORT QUESTIONS

1. State the role of hormones as chemical messengers.
2. Where are receptors located in amino acid-based hormones and steroid hormones?
3. What are neurosecretory cells?

4. Name the hormones of anterior pituitary gland.
5. Why the pituitary gland is called master gland?
6. How is the secretion of ADH controlled?
7. Write a note on parathyroid glands.
8. Write the names and functions of hormones secreted by ovaries and testes.
9. Differentiate between;
   - Exocrine and endocrine glands
   - Hypothyroidism and hyperthyroidism
   - Parathormone and calcitonin
   - Beta and alpha cells of pancreas
   - Insulin and glucagon
   - Diabetes insipidus and diabetes mellitus
   - Estrogen and progesterone
   - Positive and negative feedback

SECTION 3: LONG QUESTIONS

1. Explain the neurosecretory role of hypothalamus.
2. Describe the chemical nature of hormones.
3. State the names of the hormones secreted by pituitary gland. Write the function of each hormone and state the major abnormalities caused by the imbalance of pituitary hormones.
4. Describe the functions of thyroid hormones. What are major diseases due to their imbalance?
5. Write a detailed note on adrenal glands mentioning the hormones of adrenal context and adrenal medulla and their functions.
6. Write a note on pancreas as an endocrine gland.
7. Describe the functions of the hormones secreted by thymus glans, pineal gland, kidneys, and walls of digestive tract.

INQUISITIVE QUESTIONS

1. State the role of artificially synthesized steroids in sports and their long-term effects on its users.
2. Explain on what grounds some companies claim that growth is possible in people having short heights.

Chapter 17 HUMAN REPRODUCTIVE SYSTEM

STUDENTS'LEARNING OUTCOMES

After studying this chapter, the students will be able to:

- Describe the structures of the male reproductive system and identify their functions.
- Define male reproductive hormones and explain their functions
- Explain the structures of female reproductive system and describe their functions
- Describe the menstrual cycle and the hormones involved.

You know that reproduction means the ability of an organism to produce new offspring of its own type. In this chapter, we will learn about the human reproductive system in more detail.

17.1- Reproductive Systems

The functions of reproductive systems include the production of reproductive cells (gametes) and the preparation of gametes for fertilization. In addition, the male reproductive system delivers the gametes to the female reproductive system and the female reproductive system nourishes the fertilized egg cell and provides an environment for its development into baby.

For Information

The human reproductive system is different from other organ systems in two aspects.

i. The reproductive system is quite different in male and female. While, the other organ systems are almost the same.

ii. The reproductive system does not become functional until at puberty. In contrast, all other body systems are functional since or shortly after birth.

17.1.1- Male Reproductive System

The components of the male reproductive system and their functions are described below.

1- Gonads (Testes)

Two testes are located outside the abdomen in a skin pouch called **scrotum**. Each testis is oval-shaped and measures about 3–5 cm in length and 2–3 cm in width. A testis is composed of highly convoluted **seminiferous tubules**. The wall of the seminiferous tubules i.e., **germinal epithelium** contains two types of cells i.e.,

germline cells and **Sertoli cells**. The germinal cells make sperms (spermatozoa) by meiosis. Sertoli cell nourish the developing sperms.

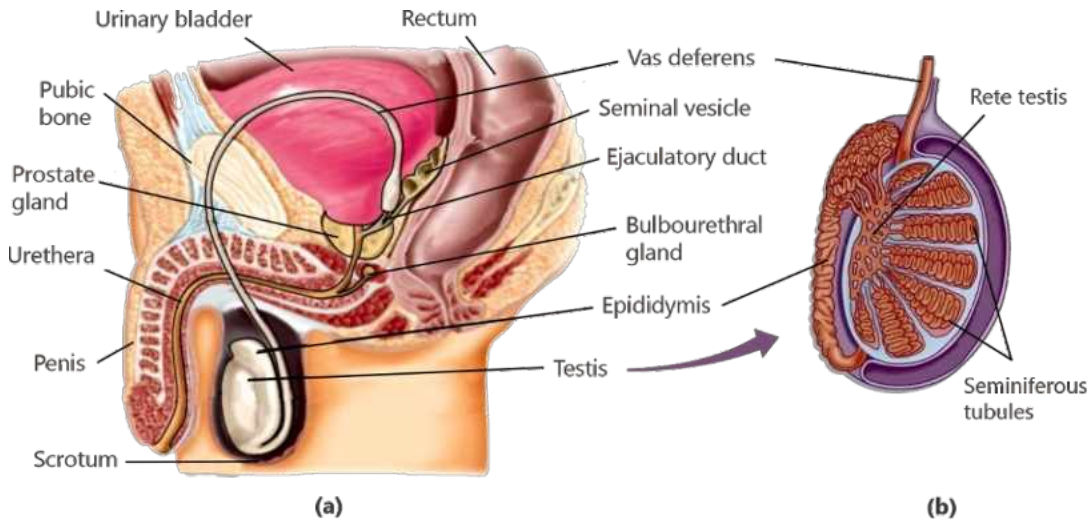


Figure 17.1: (a) Male reproductive system; (b) Cross section of a testis

2- Accessory Ducts

From seminiferous tubules, the spermatozoa enter into a network of tubules called **rete testis**. From here, they enter into a long, coiled tube called the **epididymis**. Here, the spermatozoa are still non-motile. They remain in epididymis for at least 18 hours. Then, they become motile and enter another long tube, the **vas deferens**. The vas deference passes into the abdominal cavity via the inguinal canal.

For Information

During development, the testes are formed within the abdominal cavity. Shortly before birth, they descend through an opening into the scrotum. The scrotum maintains the testes at around 34°C. This temperature is slightly lower than the core body temperature (37°C). This lower temperature is required for the development of sperms.

The vas deferens from each testis joins with a duct from a seminal vesicle (a gland). After joining, the tube is called the **ejaculatory duct**. The two ejaculatory ducts coming from two testes pass through a prostate gland. Here, they open into the urethra, which comes from the urinary bladder. The urethra opens out of the body through the penis.

3- Accessory Glands

These are the glands that add fluids to sperms to form semen.

i. There are two **seminal vesicles**. Each of these glands releases a fluid rich in fructose (sugar) into the vas deferens through a duct. This fluid provides energy for the sperm.
ii. The **prostate gland** lies just below the urinary bladder. It is soft and spongy and produces about 60% fluid of the semen. Semen is a mixture of sperms and fluids from the seminal vesicles and the prostate gland.
iii. There are two small **bulbourethral glands**. They secrete a clear fluid that lubricates the urethra for sperm passage and provides a protective neutral environment for sperms to survive.

4- Copulatory Organ (Penis)

The male copulatory organ is called penis. It contains urethra, erectile tissue, an artery and two veins.

Spermatogenesis (Production of Sperm)

Spermatogenesis is the process by which sperm are formed inside the seminiferous tubules of the testes.

In the walls of seminiferous tubules, special cells called germline cells (2n) divide by mitosis. Some of the new cells become **spermatogonia** (2n), while others remain as germline cells for future use. The spermatogonia continue to divide and form **primary spermatocytes** (2n). Primary spermatocytes move toward the inner space (lumen) of the tubules. Each primary spermatocyte divides by meiosis-I to form two **secondary spermatocytes** (1n). These cells quickly undergo meiosis-II to produce spermatids (1n). In this way, one spermatogonium (2n) produces four spermatids (1n). Each spermatid matures into an elongated sperm.

A sperm consists of three parts i.e., a head, a midpiece and a tail. The genetic material is present within its **head**. About two-thirds of the head is surrounded by a thick cap called the acrosome. The **acrosome** is formed mainly of Golgi Body. It contains enzymes that later help the sperm to fertilize the ovum. The **midpiece** contains mitochondria and a centriole. The **tail** is formed of microtubules (axial filament) which arise from the centriole of midpiece.

The mature spermatozoa present in the lumen of seminiferous tubules are non-motile. They enter the epididymis with the help of the fluid secreted by Sertoli cells. In the epididymis, they gain motility.

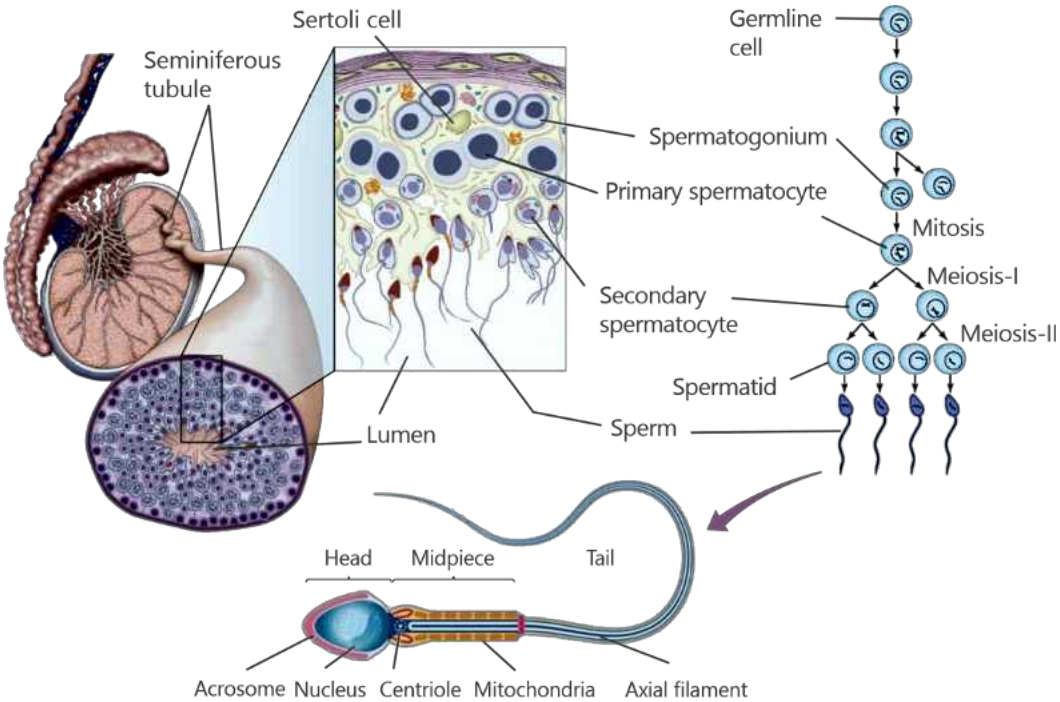


Figure 17.2: Spermatogenesis and structure of sperm

Hormonal Control of Male Reproductive System

Spermatogenesis is controlled by hormones of hypothalamus and pituitary gland. The hypothalamus releases gonadotropin-releasing hormone (GnRH). It stimulates the anterior pituitary to release follicle-stimulating hormone (FSH) and luteinizing hormone (LH).

FSH stimulates seminiferous tubules for spermatogenesis. LH stimulates **Leydig cells** (located in the interstitial tissue between the seminiferous tubules) to secrete **testosterone**.

Testosterone stimulates the development of germinal epithelium. It also maintains male secondary sexual characteristics. When FSH is released by the anterior pituitary, it causes the Sertoli cells to release a peptide hormone called

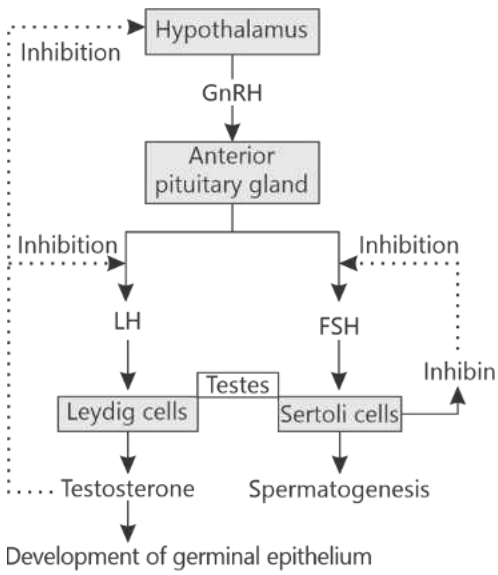


Figure 17.3: Hormonal control of male reproductive system

inhibin that inhibits further FSH secretion. Similarly, when LH stimulates Leydig cells to secrete testosterone, the release of testosterone inhibits the release of LH and GnRH.

17.1.2- Female Reproductive System

The female reproductive system consists of gonads, oviducts, uterus and vagina. The oviducts, uterus and vagina are called accessory organs of the female reproductive system.

1- Gonads (Ovaries)

Ovaries produce egg cells (ova or oocytes) and release hormones. There are two ovaries – each present on the side of the uterus. An oval-shaped ovary is 3-5 cm long and 2-3 cm wide. It contains many tiny sac-like structures called **ovarian follicles** which are the functional units of ovary. Follicles nurture immature eggs. In adult female, one of the ripening follicles ejects its egg cell each month. This event is called **ovulation**. After ovulation, the ruptured follicle is transformed into a glandular structure called the **corpus luteum**.

2- Oviducts (Fallopian tubes)

The oviducts (also called fallopian tubes or uterine tubes) are two tubes leading from the ovaries into the uterus. When an ovum matures, the ovarian follicle and ovary wall rupture to release it. The ovum enters an oviduct and travels toward the uterus. This movement is facilitated by the movements of cilia on the inner lining of oviduct. Fertilization of the ovum also occurs in oviduct. The resulting zygote undergoes several mitotic divisions while still in oviduct.

3- Uterus

The two oviducts enter into a hollow and muscular organ called uterus. It consists of three parts i.e., fundus, body and cervix. **Fundus** is the uppermost rounded portion above the openings of oviducts. The **body** of the uterus makes it major portion. The lower narrow part of uterus is called **cervix**. The opening of cervix leads to the vagina.

The wall of the uterus is composed of three layers. The **perimetrium** is the outermost thin layer. The **myometrium** is the middle thick layer composed of smooth muscles. The **endometrium** is the inner spongy lining. It has a basal layer and a functional layer. The **basal layer** has the stems cells which regenerate the functional layer. The **functional layer** thickens and is shed during the menstrual cycle. During pregnancy, it further thickens to implant the embryo. Its vascular spaces fuse and become interconnected, forming the **placenta**, which supplies oxygen and nutrition to the embryo and foetus.

4- Vagina

It is a thin-walled 8-10 cm long tube leading from the outside to the cervix of uterus. It provides a passageway for delivery of infant and for menstrual flow.

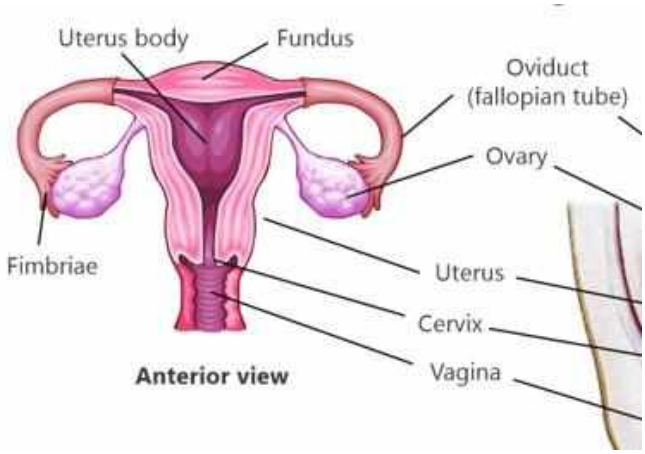


Figure 17.4: Human female reproductive system

Oogenesis (Production of Ovum)

The production of ovum starts during the embryonic development of female. In that period, the **oogonia** (2n) present in follicle divide by mitosis to produce **primary oocytes** (2n). These primary oocytes begin the first meiotic division, but stop in prophase-I. They remain in prophase-I all through childhood (for at least 10 to 14 years).

At puberty, only one of the many primary oocytes present in ovary resumes meiosis-I and produces two dissimilar haploid cells. The larger cell is the **secondary oocyte** (1n) while smaller one is called the **first polar body** (1n). The secondary oocyte stops division in metaphase-II. It released from the body. If an ovulated secondary oocyte is not fertilized by a sperm, it

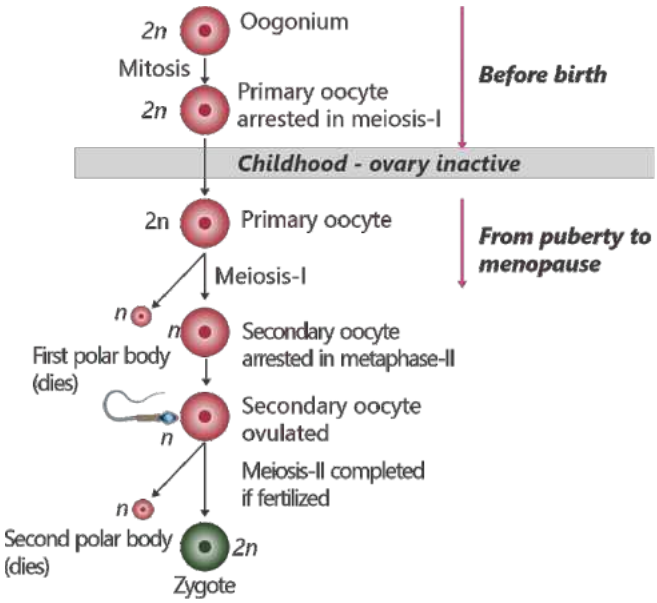


Figure 17.5: Oogenesis

dies. But, if a sperm fuses with the secondary oocyte, it quickly completes meiosis II (in oviduct). It produces two new cells i.e., one large **ovum** (1n) and a small **second polar body** (1n).

For Information
At birth, a female's ovaries contain some 2 million follicles, each with a primary oocyte.

17.1.3- Female Reproductive Cycle – The Menstrual Cycle

The female reproductive system functions by going through cyclic events, collectively called the menstrual cycle. The menstrual cycle lasts approximately one month (28 days on average). It can be divided into the following phases.

1. Menstrual Phase (Days 1-5)

In this phase, the functional layer of endometrium is shed. The detached tissue and blood move out of the body. This step is called **menstruation**. It continues for 3-5 days. It occurs due to low levels of estrogen and progesterone hormones. The gonadotropins and FSH begin to rise in this phase.

2. Proliferative Phase & Ovulation (Days 6-14)

Due to the rise in FSH, few ovarian follicles begin growing. But, only one follicle grows full and the others degenerate. This mature follicle is called **Graafian follicle** rises to the surface of the ovary.

FSH stimulates the Graafian follicle to secrete estrogen hormones. It causes the initial vascularization of endometrium and makes it velvety and thick again. As the concentration of estrogen rises, the level of FSH falls due to negative feedback. Due to the fall in FSH level, the anterior pituitary releases LH. It happens at the end of the proliferative stage (day 14). The sudden release of LH from the anterior pituitary stimulates Graafian follicle to release the ovum into the oviduct. This phase is known as **ovulation**.

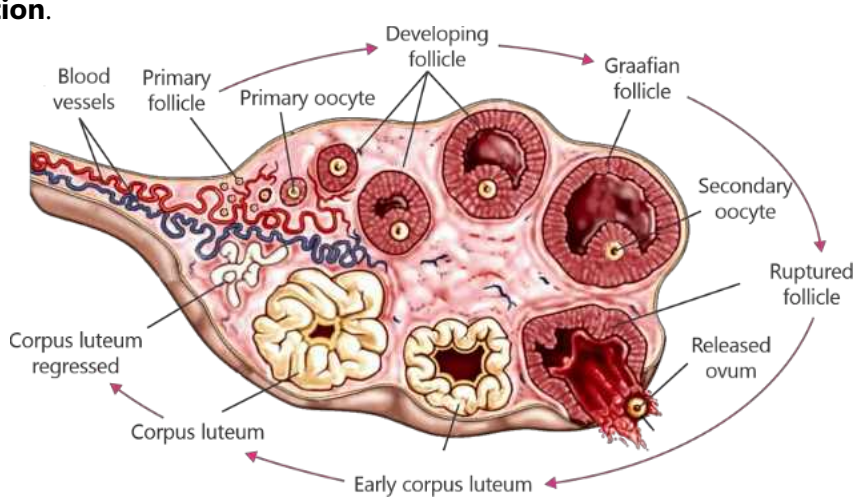


Figure 17.6: Changes in ovary during menstrual cycle

3. Secretory/Post-Ovulatory Phase (Days 15-28)

After ovulation, LH stimulates the ruptured Graafian follicle to develop into a yellow structure called the **corpus luteum**. Copus luteum secretes estrogen and progesterone hormones which make the endometrium more vascular and glandular.

For Information
In human females, menstrual cycle stops around 50 year of age and it is termed as menopause.

If fertilization does not occur, the corpus luteum starts to degenerate. As a result, the anterior pituitary stops secreting LH. The destruction of corpus luteum also results in an abrupt fall in the concentration of estrogen and progesterone. It causes the thickened endometrium to be shed with bleeding i.e., menstruation on day 28.

For Information
When the ovulated oocyte is fertilized, the tiny embryo secretes human chorionic gonadotropin (hCG). This hormone maintains the corpus luteum. So, the levels of estrogen and progesterone keep high and menstruation is prevented. Because hCG comes from the embryonic chorion and not the mother, it is the hormone that is tested for in all pregnancy tests.

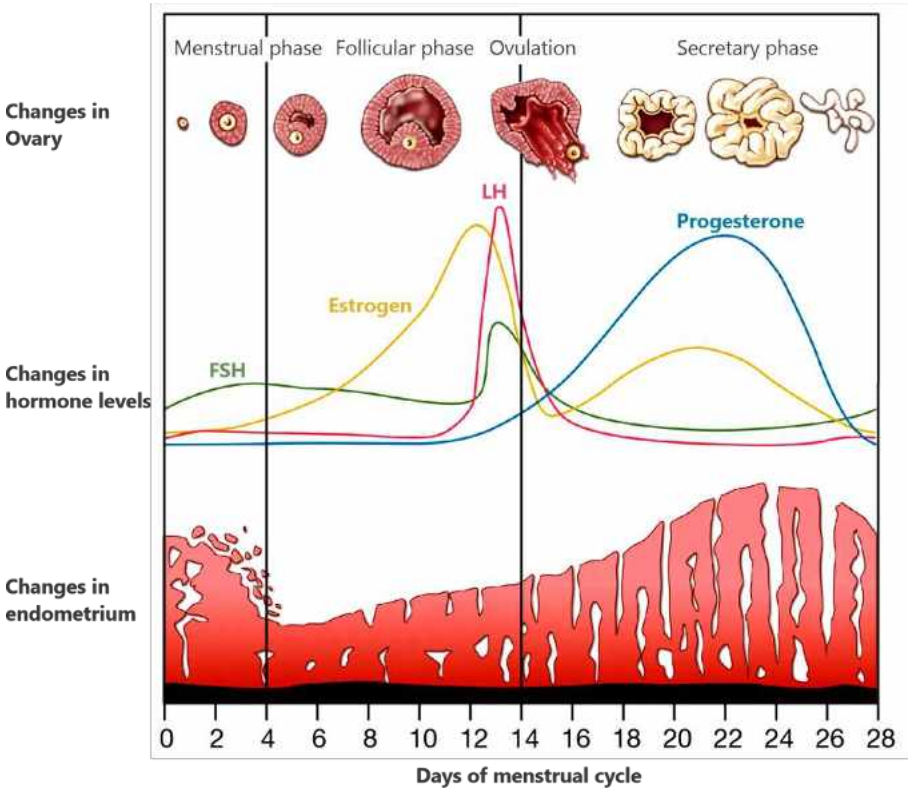


Figure 17.7: Menstrual cycle

EXERCISE

SECTION 1: MULTIPLE CHOICE QUESTIONS

1. Fertilization of the ovum occurs in:
   (a) Oviduct (b) Uterus (c) Cervix (d) Vagina
2. Which cells of the testis secrete testosterone hormone?
   (a) Sertoli cells (b) Germline cells (c) Spermatogonia (d) Leydig cells
3. The site of the production of sperm is:
   (a) Seminiferous tubules (b) Epididymis
   (c) Seminal vesicle (d) Vas deference
4. Which part produces fructose-rich fluid for the sperm?
   (a) Prostate gland (b) Seminal vesicles
   (c) Testis (d) Bulbourethral gland
5. In males, which hormone initiates spermatogenesis?
   (a) Luteinizing hormone b) Follicle stimulating hormone
   (c) Gonadotropin-releasing hormone (d) Testosterone
6. During ovulation which cell is actually released from ovary?
   (a) Oogonium (b) Primary oocyte
   (c) Secondary oocyte (d) Mature ovum
7. Which hormone stimulates the Graafian follicle to release ovum?
   (a) LH (b) FSH
   (c) Estrogen (d) Progesterone

SECTION 2: SHORT QUESTIONS

1. Briefly describe the function of seminal vesicles, prostate gland, and bulbourethral glands.
2. When do the ova in a females begin meiosis? When do they complete the first meiotic division?
3. Write a brief note on the role of FSH, LH and testosterone in man.
4. List the structures of female reproductive system.
5. What changes occur in ovulation and menstruation during gestation period.
6. What is the role of the corpus luteum in a menstrual cycle?
7. Differentiate between:
   - Primary and secondary spermatocytes
   - Spermatogenesis and oogenesis
   - Sertoli cell and Leydig cell
   - Spermatids and spermatozoa

- Primary oocyte and secondary oocyte

SECTION 3: LONG QUESTIONS

1. Describe the structure of human male reproductive system. Identify the function of each part.
2. Explain the major reproductive hormones of human male and explain their role.
3. Explain the structures of human female reproductive system. Identify the function of each part.
4. Describe the events of a menstrual cycle and explain its hormonal regulation.

INQUISITIVE QUESTIONS

1. List the structures in order, through which a sperm passes on its way from the seminiferous tubules to the outside.
2. Describe the effect of endocrine disrupting contaminants on the reproductive abilities.
3. Why are so many sperms produced in the male and so few ova produced in the female?

Chapter 18

INHERITANCE

STUDENTS'LEARNING OUTCOMES

After studying this chapter, the students will be able to:

- Define gene.
- Explain the law of segregation and independent assortment, using a suitable example related to the pea plants.
- Relate the Law of independent assortment to random orientation of chromosomes during meiosis.
- Express limitations of independent assortment and its usefulness.
- Show that independent assortment leads to variation in the gametes.
- Evaluate that inheritance of genes and their mixing during fertilization is based on mathematical probabilities.
- Describe the exceptions to the Mendel's laws of inheritance.
- Explain incomplete dominance and exemplify it through the inheritance of flower color in 4 O' clock plant.
- Differentiate between incomplete dominance and codominance.
- Define alleles and multiple alleles.
- State the alleles responsible for the trait of ABO blood groups.
- Explain the case where two alleles have equal dominance through the genetics of human blood group AB.
- Name the various human blood group systems.
- Investigate the reasons for O-ve individual as the Universal donor and AB +ve as the Universal recipient.
- Describe the occurrence of some other blood group systems.
- Associate the positive and negative blood groups with the presence and absence of Rh factor.
- Justify why Rh incompatibility could be a danger to the developing fetus and mother.
- Explain Erythroblastosis fetalis in the light of antigen-antibody reaction.
- Suggest measures to counter the problem of Erythroblastosis fetalis before it occurs.
- Define and relate the terms; polygenic and epistasis.
- Describe polygenic inheritance using suitable examples from plants (grain color in wheat) and animals (skin color in man).
- List at least five polygenic traits discovered in humans.
- Give one example of epistasis from mammals (coat color inheritance in Labrador retrievers) and one from plants (pigment phenotype in foxgloves) and justify modified Mendelian ratios.
- Describe the terms gene linkage and crossing over.
- Explain that gene linkage counters independent assortment and crossing-over modifies the progeny.

- Suggest that linkage can be observed / evaluated only if the number of progeny is quite large.
- Explain the XX-XY mechanism of sex determination in mammals.
- Identify male and female individuals from the karyotype of man.
- Solve the genetics problems related to XX-XY, sex determination.
- Describe the concept of sex-linkage.
- Explain the inheritance of sex-linked traits (eye color) in Drosophila.
- Describe the sex-linked inheritance of male characters due to Y-chromosome and the effect of Holandric genes.
- Describe the X-linked disorders with reference to the patterns of inheritance.
- Name some of the sex-linked disorders of man (Red green color blindness, Hemophilia) .
- Explain the techniques employed for embryonic screening e.g., Amniocentesis and Chorionic Villus Sampling.

You have studied the basics of Mendelian genetics in Grade X. This chapter carries the concept forward to post-mendelian research. It also gives an insight into the inherited diseases and their symptoms and treatment.

Recalling

The Austrian monk, Gregor Johann Mendel, was the first to explain the mechanism of inheritance. He developed true-breeding (pure-breeding) varieties of pea plants. A plant which is true-breeding for a particular trait produces all the offspring with the same trait upon self-fertilization.

18.1- Law of Segregation

Mendel performed monohybrid crossed in pea plant. In the first monohybrid crosses, he observed the inheritance of "seed shape". When he crossed a true-breeding round-seeded plant with wrinkled-seeded plant, all plants of F1 generation produced round seeds. He called the trait that expressed in the F1 plants (round seeds) as **dominant** and the alternative trait that was not expressed in the F1 plants (wrinkled seeds) as **recessive** trait.

When F1 plants were self-pollinated, the 1/4 of F2 seed were wrinkled. In other words, the dominant-recessive ratio among the F2 seeds was 3 dominant : 1 recessive. His results suggested that the 3:1 ratio in F2 generation was really a disguised 1:2:1 ratio.

In F2 generation,

- 1⁄4 were pure-breeding dominant
- 2⁄4 were not pure-breeding dominant
- 1⁄4 were pure-breeding recessive

From his experiments, Mendel concluded that for each pair of alternative traits, one trait was not expressed in the F1 hybrids, although it reappeared in 1/4 of F2 seeds. The trait that "disappeared" must therefore be disguised in the F1 seeds.

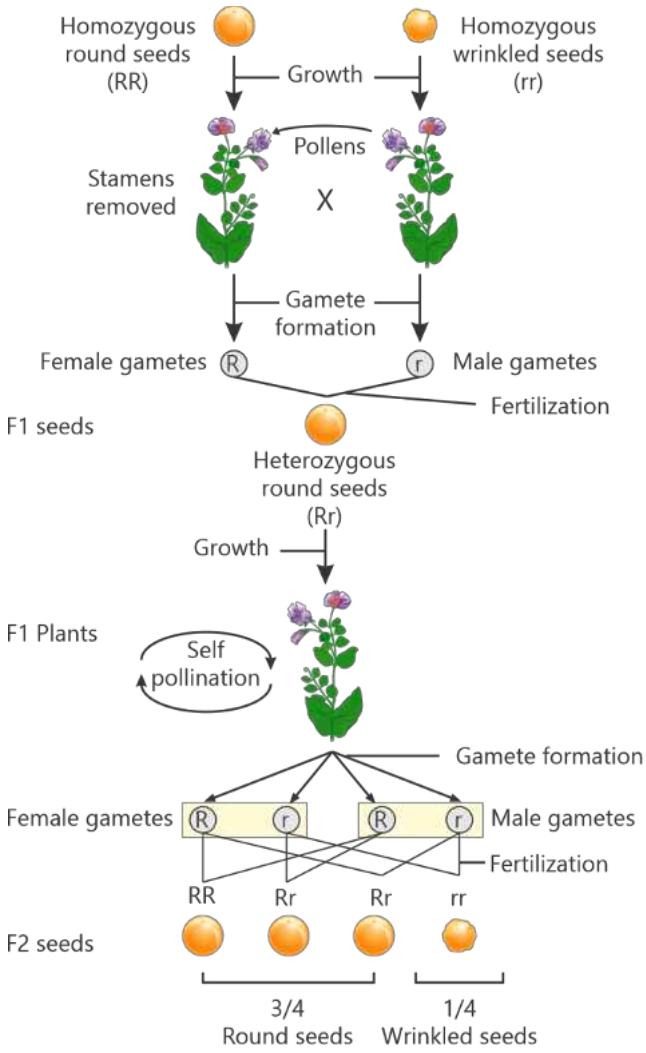


Figure 18.1: Mendel's monohybrid cross

Interpretation of the Results

Mendel explained that the characters (traits) of organisms are controlled by special "factors" (which are now called **genes**). Each individual is diploid and has two genes for a character. Gene are present on chromosomes. When the individual forms haploid gametes (eggs or sperms), each gamete receives one gene for each character.

All copies of a gene may not be identical. The alternative forms of a gene, leading to alternative traits, are called **alleles**. When two haploid gametes containing

the same alleles fuse, the offspring receives the gene-pair with similar alleles. Such offspring is called **homozygous** for that gene pair or trait. When the two haploid gametes with different alleles fuse, the offspring is **heterozygous**.

We now know that a gene is **"a sequence of nucleotides as part of DNA, which codes for the formation of a polypeptide"**. The particular location of a gene on a chromosome is called the gene's locus.

When the individual forms gametes, each gamete receives just one gene copy (allele), which is selected randomly. This is known as the law of segregation. When gametes join, they form a new individual, whose genotype consists of the alleles contained in the gametes.

For Information

In heterozygous individuals, only one allele (dominant) is expressed, while the other allele (recessive) is present but unexpressed.

The combination of alleles in an individual is its **genotype**. The **phenotype** is the observable character or trait. In other words, the genotype is the blueprint, and the phenotype is the visible outcome.

Testcross

A testcross is used to determine whether a dominant phenotype is homozygous or heterozygous. It involves crossing the dominant phenotype with a recessive phenotype. For example;

A round-seeded plant may be RR (homozygous) or Rr (heterozygous), while wrinkled-seeded plants are always rr. To determine its genotype, the round-seeded plant is crossed with a wrinkled-seeded plant (rr).

- If it is **RR**, all offspring will be **round (Rr)**.
- If it is **Rr**, the offspring will be **round and wrinkled in a 1:1 ratio**.

18.2- Law of Independent Assortment

After showing that alleles of one character segregate independently, Mendel studied whether genes of different characters also segregate independently. He examined two characters together: seed shape — round (R) dominant and wrinkled (r) recessive, and seed colour — yellow (Y) dominant and green (y) recessive. He established pure-breeding lines for these traits.

1. Pure-breeding plants with both dominant traits i.e., round and yellow seeds (RRYY)
2. Pure-breeding plants with both recessive traits i.e., wrinkled and green seeds (rryy)

He crossed pure-breeding plants (**RRYY** × **rryy**). All F1 seeds showed both dominant traits (round and yellow) and were heterozygous with genotype **RrYy**. These F1 plants are called **dihybrids**, as they are heterozygous for both genes.

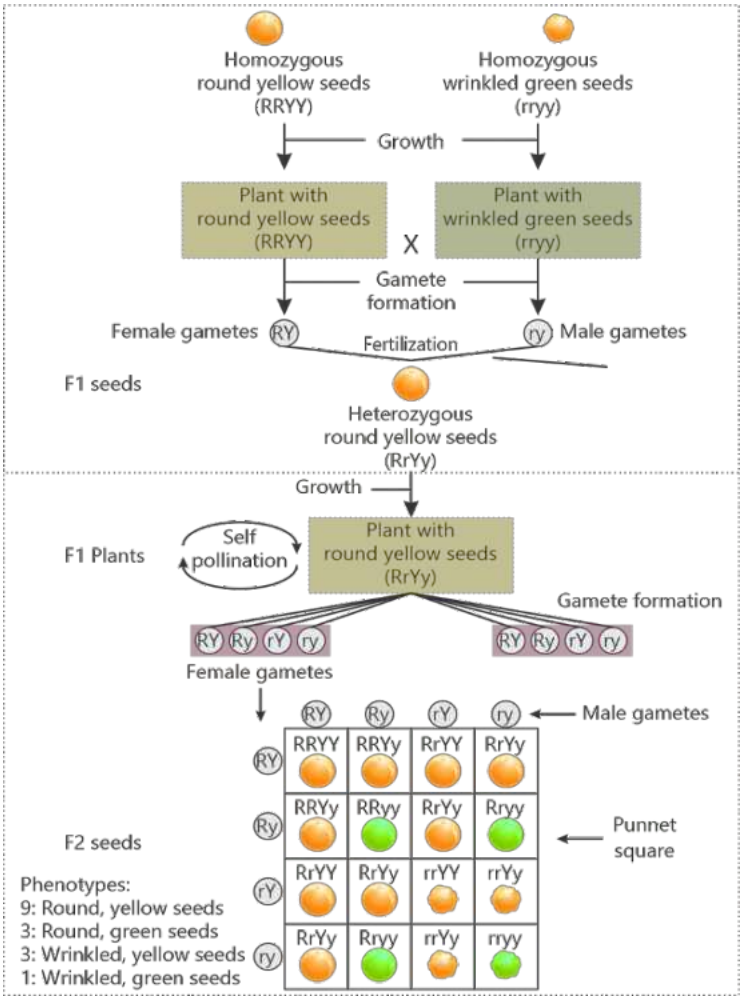


Figure 18.2: Mendel's dihybrid cross

To confirm the genotype of F1 plants, Mendel self-fertilized them to produce the F2 generation. Instead of a 3:1 ratio, the F2 showed four phenotypes i.e., round yellow, round green, wrinkled yellow, and wrinkled green - in a **9:3:3:1 ratio**.

For Information

Independent assortment of different genes does not affect the segregation of individual allele pairs. Thus, round vs wrinkled seeds show a 3:1 ratio, and yellow vs green seeds also show a 3:1 ratio.

Interpretations of the Results

In a F1 dihybrid plant (**RrYy**), seed shape has alleles **R, r** and seed colour has **Y, y**. During gamete formation, four combinations are produced: **RY, Ry, rY, ry**. When these gametes combine (4 male × 4 female gametes), **16 combinations** are formed in the F2 generation. This results in four phenotypes in F2 in a **9:3:3:1 ratio**:

- 9 round yellow
- 3 round green
- 3 wrinkled yellow
- 1 wrinkled green

The results showed that genes for seed shape assort independently of genes for seed colour. On these basis, Mendel proposed that "each pair of alleles assorts independently of other pairs of alleles during gamete formation". It is referred to as Mendel's law of independent assortment.

Independent Assortment as a Source of Variations

Independent assortment of alleles creates genetic diversity. It mixes alleles during the formation of gametes (sperm and egg cells). Durin gamete formation, alleles for different traits, which are on different chromosomes, separate independently of each other. This means one character (like eye colour) does not affect another character (like hair texture).

Tidbit
A modern re-statement of Mendel's law of independent assortment would be that *"genes that are located on different chromosomes assort independently during meiosis"*.

Independent assortment happens because homologous chromosome pairs line up randomly during meiosis. This random alignment causes different combinations of maternal and paternal alleles in gametes. In humans, it can produce over 8 million (2^{23}) possible allele combinations in one gamete. This reshuffling makes every gamete genetically unique.

The chances of variation increases further during fertilization. When a sperm and egg fuse, they combine their unique alleles to form a genetically unique zygote. This continuous mixing of alleles in a population creates genetic diversity, which helps species adapt to changing environments.

18.2.1- Limitations to the Law of Independent Assortment

The law of independent assortment helps explain how genes are inherited, but it does not cover all genetic complexities. For example;

1. **Linked genes:** Genes close together on the same chromosome are often inherited together, not independently. This phenomenon is known as **gene linkage**.
2. **Chromosome crossover:** It is the exchange of genetic material between homologous chromosomes during meiosis. Due to this process, the ratio of gene combinations may not be as predicted by the law of independent assortment.
3. **Epistasis:** It is phenomenon where the effect of one gene is modified or suppressed by one or more unrelated genes Epistasis also changes the expected ratio of gene combinations.

4. **Pleiotropy:** It is the phenomenon where one gene controls multiple characters. It causes the characters to be linked and controlled by a single gene. Pleiotropy can distort the expected phenotypic ratios from independent assortment.
5. **Environmental effect:** Many phenotypes can be affected by the environment.

18.2.2- Inheritance and Mathematical Probabilities

The transfer of genes from one generation to the other is random. The calculation of the possible new allelic combinations is based on mathematical probability. Probability means how often a particular outcome occurs.

Probability of inheritance of one or two alleles (e.g., RrYy × RrYy) can be calculated using a Punnett square. However, for many genes (e.g., AaBbCcDdEe × AaBbCcDdEe), Punnett squares become too complex. So, probability rules are used instead.

Basic Rule of Probability

Probability is calculated by dividing the number of times an event occurs by the total number of possible outcomes.

For example, if wrinkled pea seeds appear 1,850 times out of 7,324 seeds, the probability is 1,850 ÷ 7,324 = 0.253, which is about 1 in 4 seeds.

Performing and Recoding
Use the rules of probability and calculate the probability of getting Sixes when you roll a dice 100 times.

The Product Rule of Probability

The product rule of probability says that the chance of two or more independent events happening together is calculated by multiplying their individual probabilities. For example, getting a six on one dice is 1/6. For two dice, the chance of getting two sixes is (1/6) × (1/6) = 1/36.

The product rule can be used to predict fertilization outcomes.

Example 1 (Aa × Aa):

- Each parent has a 1/2 chance of giving gamete with allele *a*.
- So, probability of aa offspring = (1/2) × (1/2) = 1/4.

Example 2 (RrYy × RrYy):

- Probability of Round seed = 3/4, Probability of yellow seed = 3/4.
- So, probability of round yellow offspring = (3/4) × (3/4) = 9/16.

The Sum Rule of Probability

This rule applies to mutually exclusive events (only one can happen at a time). The probability of either event is found by adding event's individual probabilities. For example, on a six-sided dice, the chance of getting 1 is 1/6 and the chance of getting 6 is 1/6. So, getting either 1 or 6 will be (1/6) + (1/6) = 1/3.

Example

In a cross **Aa × Aa**, dominant phenotype (AA or Aa) can occur in three ways:

- A from male + A from female (AA)

- A from male + a from female (Aa)
- a from male + A from female (Aa)

Each individual event has a probability of **1/4**.

So, total probability of dominant phenotype = (1/4) + (1/4) + (1/4) = **3/4**.

Analyzing, Interpreting and Communication

Solve the following genetic problems.

- Phenylketonuria and albinism are two recessive disorders which assort independently. If a mother and a father who both are heterozygous for both traits, produce a child, what is the chance of their having a child with (a) phenylketonuria, (b) albinism, and (c) both traits?
- A gene has two alleles i.e., dominant C and recessive c. What proportions of the offspring from a CC x Cc cross would be homozygous dominant, homozygous recessive and heterozygous?
- A TtYy pea plant self-pollinates and one seed is picked at random for planting. (a) What is the chance that the seed will produce a tall, green seeded plant? (b) If it turns out to be tall and yellow-seeded what is the chance that its genotype is TTYY?

18.3- Dominance Relations and Multiple Alleles

In pea plants (Yy), the dominant allele (Y) is fully expressed and the recessive allele (y) is not expressed at all. So no intermediate trait (green) appears in Yy. This is called **complete dominance**. Scientists tested Mendel's ideas and found that results were not always the same. Many traits do not show complete dominance. Moreover, some traits are controlled by more than two alleles.

18.3.1- Incomplete Dominance

In **incomplete dominance**, neither allele is fully dominant over the other. In such situations, the heterozygous individual shows a phenotype that is intermediate between phenotypes of both parents. For example, in Japanese four-o'clock plants, when a red flowered (RR) is crossed with white flowered (WW), it produces all heterozygous pink flowered (RW) offspring. This shows neither the alleles for red (R) nor of white (W) is completely dominant. When two pink flowered

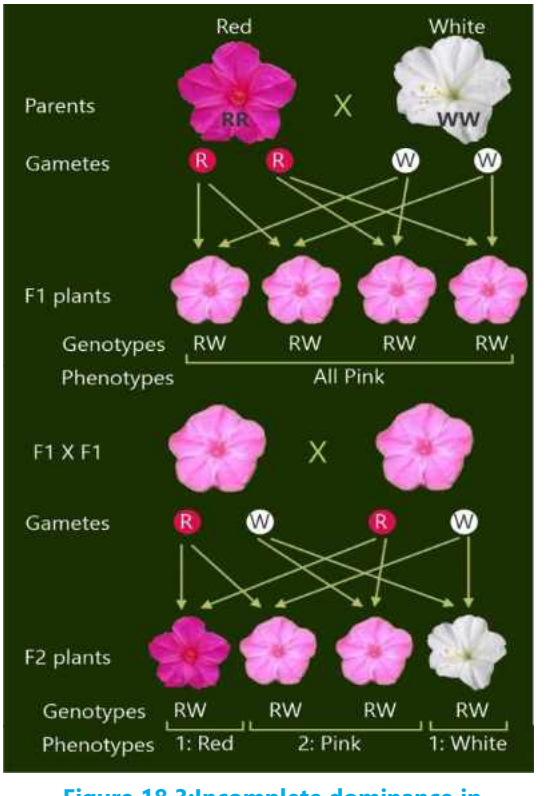


Figure 18.3:Incomplete dominance in Japanese 4-o'clock plants

plants (RW × RW) are crossed, the result is red (RR), pink (RW), and white (WW) in a **1:2:1 ratio**.

Solve the following genetic problem:
A pink flower four O'clock plant is crossed with a red flower plant. Find:
(a) Probability of Red flower plant
(b) Probability of pink flower plant
(c) Ratio of pink flower to red flower plant

18.3.2- Co-dominance

Co-dominance is type of inheritance in which both alleles in a gene pair are fully expressed in a heterozygous individual. In co-dominance, both alleles work without blending or one masking the other. So, the products of both alleles appear together in the phenotype of heterozygous.

For Information
The "L" stands for Landsteiner and Levine, the scientists who discovered this system in 1927.

For Information
It is also called the MNS system because the genes of MN group and the S/s group are physically "linked" on the same chromosome.

The human **MN blood group** shows codominance, where both alleles are equally expressed. There are two alleles for this system.

- Allele L^M Produces M antigen.
- Allele L^N Produces N antigen.

Both L^M and L^N are codominant. So, if both are present, both antigens appear on red blood cells. In this way, there are three possibilities i.e.,

1. M blood group (L^ML^M) – individuals with only M antigens.
2. N blood group (L^NL^N) – individuals with only N antigens.
3. MN blood group (L^ML^N) – individuals with both M and N antigens.

MN Blood Group System

Genotype	Antigen	Blood Group Phenotype
L^ML^M	M	M blood group
L^NL^N	N	N blood group
L^ML^N	M & N	MN blood group

Table: Comparison between Incomplete Dominance and Codominance

Incomplete dominance	Co-dominance
1. In heterozygous state, both genes blend their phenotypic effects.	1. In heterozygous, both genes fully express their effects.
2. The heterozygous show an intermediate phenotype between the two parental phenotypes.	2. The heterozygous show both parental phenotypes at a time.

3. Example: Flower colour in Japanese 4 O' clock plant	3. Example: Human MN blood group and human AB blood group

18.3.3- Multiple Alleles

A **multiple allelic system** occurs when a gene has more than two alternative forms (alleles) in a population. This creates more variation in traits.

An example is the human **ABO blood group system**. It is controlled by one gene (I gene) with three alleles: **I^A, I^B, and i**. Each person has only two of these alleles. Different combinations produce four blood groups:

Phenotypes (Blood groups)	Genotypes
1. **A**	I^AI^A or I^Ai
2. **B**	I^BI^B or I^Bi
3. **AB**	I^AI^B
4. **O**	ii

For Information
In human ABO blood group system, the alleles I^A and I^B also show co-dominance.

18.4- Blood Group Systems

A blood group system is a way of classifying blood based on the presence of specific antigens on red blood cells (RBCs). The International Society of Blood Transfusion recognizes **43 blood group systems** in humans. Each system has different blood groups.

For Information
In blood group systems, the antigens present on the surface of RBCs may be proteins, carbohydrates, glycoproteins, or glycolipids.

The most important is the **ABO system** (blood groups A, B, AB, O), and the second is the **Rh system** (blood groups Rh-positive and Rh-negative). For blood transfusion, the matching of ABO and Rh-system is necessary between donor and recipient's blood. The following are the other major blood group system, but these usually do not cause matching problems in the blood transfusions.

Blood Group System	Well-known antigens
MNS	M, N, S, s, U
Lutheran	Lu^a and Lu^b
Kell	K, k, Kp^a, Kp^b, Js^a, Js^b
Lewis system	Le^a and le^b
Duffy	Fy^a, Fy^b, Fy^3

18.4.1- ABO Blood Group System

The ABO system was discovered by Landsteiner in 1901. He found two antigens on red blood cells (RBCs): **antigen-A and antigen-B**. He observed some individuals have antigen-A on their RBCs while others have antigen-B. He also observed that many individuals do not have any of these antigens. Later, Landsteiner's students discovered

that some individuals have both A and B antigens on their RBCs. So, they named the different blood groups as:

- People with antigen-A — blood group A
- People with antigen-B — blood group B
- People with both (antigen- A & antigen-B — blood group AB
- People with neither of antigen-A & antigen-B — blood group O

Each person's blood plasma contains antibodies against the antigen s/he does not have:

- Blood group A → anti-B antibodies
- Blood group B → anti-A antibodies

This is called **Landsteiner's rule of antigen–antibody relationship**.

For Information
Some people secrete antigens A and B in body fluids e.g., saliva, tears, urine. They possess a dominant secretor gene "Se" on their chromosome 19.

Inheritance of the ABO Blood Groups

The ABO system is controlled by the gene I ("I" stands for iso-haem-agglutinogen). It is located on chromosome 9 and has multiple alleles i.e., I^A, I^B and i. The alleles I^A and I^B are co-dominant over each other. While allele i is recessive to both of them. The I^A allele produces antigen-A, the I^B allele produces antigen-B. Allele i does not produce any antigen. Therefore;

- Genotypes I^AI^A and I^Ai produce antigen-A. This phenotype is called **blood group A**. Individual's plasma contains anti-B antibodies.
- Genotypes I^BI^B and I^Bi produce antigen-B. This phenotype is called **blood group B**. Individual's plasma contains anti-A antibodies.
- Genotype I^AI^B produces both antigen-A and antigen-B. This phenotype is called **blood group AB**. Individual's plasma does not contain anti-A or anti-B antibodies.
- Genotype ii produces neither antigen-A nor antigen-B. This phenotype is called **blood group O**. Individual's plasma contains both anti-A and anti-B antibodies.

For Information
In ABO system, the production of anti-A and anti-B antibodies does not require any exposure to antigens. Rather, these antibodies start forming from embryonic life and continue throughout life.

Solve the following genetic problem.

- A person has blood group A while his wife has B. They have four children each with different blood group. Explain that how is it possible?
- A woman with blood group B has a child with blood group O. What are the genotypes of the mother and child?

For Information
In pea plants, each character that Mendel studied was controlled by a gene with two alleles. One allele was completely dominant over the other. But incomplete dominance and co-dominance do not obey this rule of dominance. Similarly, in multiple allelic system, a gene has more than two alleles,

Which genotypes could the father not have?

any two of which work for producing a phenotype.

Genotypes	I^AI^A or I^Ai	I^BI^B or I^Bi	I^AI^B	ii
Antigens (on RBCs)	Antigen-A	Antigen-B	Antigen-B Antigen-A	No Antigens
Antibodies (in Plasma)	Anti-B	Anti-A	No Anti-A antibody No Anti-B antibody	Anti-A Anti-B
Phenotypes (Blood Groups)	A	B	AB	O

Figure 18.4: ABO system genotypes and phenotypes

Principle of Transfusion in ABO System

During blood transfusions between two incompatible (unmatching) blood groups, the recipient's antibodies attack the antigens on donor's RBCs. It causing **clumping (agglutination)** of RBCs. It happens;

- If blood group A individual receives blood group B and vice versa, and
- If donor's blood group is AB and the recipient is A, B, or O.

Exceptions;

- O blood group (donor): has no antigen-A and antigen-B. So, it can be given to A, B, AB, or O. So, O blood group are called **universal donors of ABO system**.
- AB blood group (recipient): has no anti-A or anti-B antibodies. So, it can receive all blood types. So, AB group are called **universal recipients of ABO system.**

Justify why a recessive blood group allele 'i' is more frequent in the population.

- In a cross between blood group O (ii) and O (ii), all offspring will have blood group O (ii).
- In a cross between heterozygous I^Ai and I^Ai, 25% offspring will have blood group O (ii)
- In a cross between heterozygous I^Bi and I^Bi, 25% offspring will have blood group O (ii).
- In a cross between heterozygous I^Ai and I^Bi, 25% offspring will have blood group O (ii).

That's why blood group allele 'i' is more frequent in the population.

18.4.2- Rh Blood Group System

It is the second most important blood group system in humans. The name of this system (Rh) is derived from **Rhesus monkey**, in which its antigen was first discovered by Landsteiner in 1930s. This system is based on the presence or absence

of **Rh-antigen** (also called Rh factor or antigen-D) on RBCs. The individuals having this antigen are called **Rh-positive** and those in which it is absent are called **Rh-negative**.

Usually, the blood group of an individual mentions both ABO and Rh systems together. For instance, the individual who has antigen-A and Rh-antigen will have blood group A-positive. The individual who has antigen-A but lack Rh-antigen will have blood group A-negative. Similar is the case with individuals of blood groups B, AB and O.

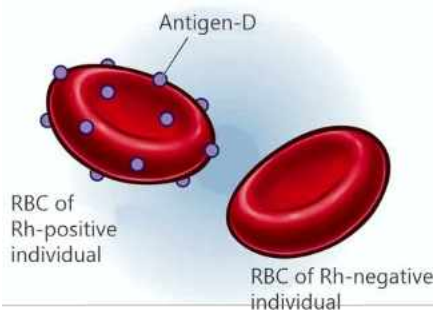


Figure 18.5: The Rh-blood group system

Inheritance of Rh Blood Groups

Rh blood group system is controlled a gene "D". It has two alleles (D and d). The dominant allele D forms the Rh-antigen, while the recessive allele d inhibits the formation of Rh-antigen. Therefore, individuals with genotypes DD or Dd have Rh-antigen and are Rh-positive. While the individuals with genotype dd do not have Rh-antigen and are Rh-negative.

Principle of Transfusion in Rh System

In ABO blood groups, the anti-A or anti-B antibodies are already present in plasma. But in Rh system, Rh-negative people do not naturally have anti-Rh antibodies. But, if Rh-negative person receives Rh-positive blood, his body start producing anti-Rh antibodies, which destroy the donated RBCs. These antibodies stay in the body for life.

The blood group O-negative is the **actual universal donor** because neither has any antigen of ABO system nor has antigen of Rh system. The blood group AB-positive is the **actual universal recipient** because it has neither anti-A and anti-B nor anti-Rh antibodies. Therefore, it receives any donor's blood.

18.4.3- Erythroblastosis Foetalis

Erythroblastosis foetalis is a **haemolytic disease** of the foetus/new-born caused by **Rh incompatibility** between Rh-negative pregnant mother and Rh-positive foetus.

Reason and Complications

It occurs when Rh-negative mother (dd) carries Rh-positive baby (DD or Dd). Foetal blood may enter the mother's blood. On exposure to Rh-antigens of feotus, mother will produce anti-Rh antibodies. These antibodies can cross the placenta and enter foetus blood. Here, mother's and anti-Rh antibodies will destroy foetal RBCs, leading to **haemolytic anaemia** and other serious complications in foetus.

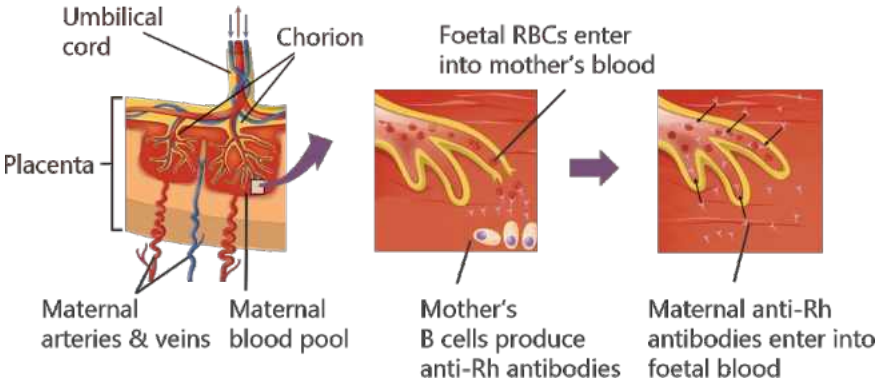


Figure 18.6: Erythroblastosis foetalis

In severe cases, destruction of foetal RBCs can cause **miscarriage or stillbirth**. If the foetus survives, the liver and spleen become enlarged because they try to make more RBCs. The breakdown of RBCs produces bilirubin, which builds up and causes jaundice (yellow skin and eyes) in foetus. High bilirubin can also damage brain cells. The surviving baby's blood is replaced by Rh-negative blood free of anti-Rh antibodies.

Tidbit

Sometimes ABO incompatibility protects the baby against Rh incompatibility. If O-negative mother conceives A-positive or B-positive baby, any foetal RBCs entering the mother's blood are quickly destroyed by her already present anti-A or anti-B antibodies before she can form anti-Rh antibodies.

Tidbit

When there are chances of Rh-incompatibility, the mother is given an injection of Rh-antiserum (serum containing anti-Rh antibodies) during early pregnancy and immediately after birth. This causes the destruction of any of the baby's RBCs that may have crossed into mother's blood before sensitizing the mother's immune system.

Solve the following genetic problem.

An Rh-negative woman is married to an Rh-positive man, whose father was also Rh-negative. What are the possible genotypes of each person in the family, what are the chances that their child will be affected with erythroblastosis foetalis?

18.5- Polygenic Inheritance and Epistasis

Polygenic inheritance and epistasis are further examples of deviation from Mendel's inheritance.

18.5.1- Polygenic Inheritance

Polygenic characters are controlled by many gene pairs, not just one. In polygenic inheritance, all alleles of many genes contribute to the same trait in an **additive way** (some increase, some decrease the effect). The combined effect produces a single trait.

Polygenic traits show a range of differences i.e., **continuous variation** in a population. Examples of polygenic traits include height, skin colour, weight, intelligence in humans, and grain colour in plants.

Inheritance of Human Skin Colour

Skin colour depends on the amount of **melanin** pigment. More melanin means darker skin; less melanin means lighter skin. This trait is controlled by polygenes (about 60 genes), but for understanding, we take 3 genes: **A, B, C**. Each of these genes has a dominant and a recessive allele.

- Alleles **A, B, and C** produce more melanin
- Alleles **a, b, and c** produce less melanin

If the parents have genotypes AABBCC (very dark) and aabbcc (very light), all of their children will be AaBbCc (intermediate colour). If both parents are triple heterozygotes i.e., AaBbCc, their children will have wide range of skin colours from very dark to very light. The ratio of different skin colours will be **1 : 6 : 15 : 20 : 15 : 6 : 1** (from very dark to very light).

- **AABBCC** → darkest (maximum melanin)
- **aabbcc** → lightest (minimum melanin)

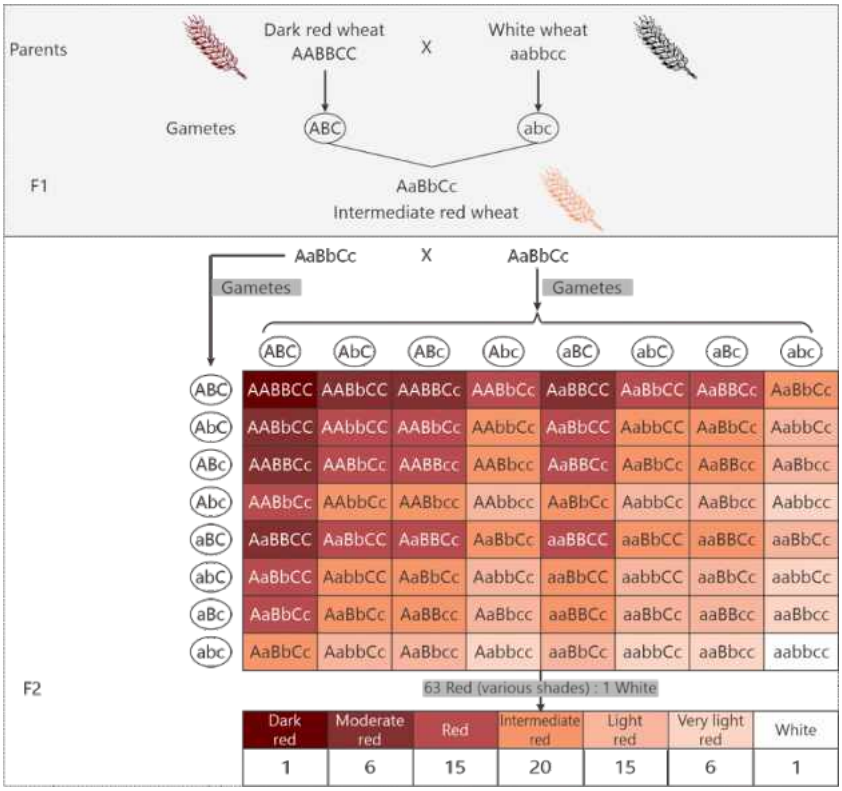


Figure 18.7: Polygenic inheritance of what grain colour

Wheat Grain Colour

Wheat grain colour shows continuous variation, ranging from white to dark red. This trait is controlled by three polygenes: A, B, and C. Each gene has a dominant allele (A, B, C) that produces red pigment and a recessive allele (a, b, c) that does not. The genotype AABBCC produces the maximum red pigment, so the grains are dark red. In contrast, aabbcc produces no pigment, so the grains are white. See the following cross.

When dark red wheat (AABBCC) is crossed with white wheat (aabbcc), the F1 generation (AaBbCc) shows an intermediate red colour. In the F2 generation, during crossbreeding, a wide range of colours appears. Only one plant will darkest red grains (AABBCC) and one will plant will have white grains. While 62 plants will show different shades of red (moderate red to very light red).

For Information
Environmental factors like light, water and nutrients also influence the grain colour.

18.5.2- Epistasis

Most traits appear due to interactions between different genes. These interactions are of two main types.

1. In **allelic interactions**, alleles of the same gene interact with each other. Examples include incomplete dominance, codominance, and multiple alleles.

2. In **non-allelic interactions**, alleles of different genes (at different loci) interact with each other. A common example of this type is epistasis.

Epistasis is defined as the gene interaction, in which an allele masks or modifies the effect of the alleles of other genes (present at other loci). The allele which masks or modifies the effect is called **epistatic**. While, the allele whose effect is masked or modified is called **hypostatic**. For example, in humans, the allele of baldness is epistatic to the alleles of black and blonde hair.

For Information
Both polygenic inheritance and epistasis involve more than one gene, but they work differently.
In polygenic inheritance, many genes act together, each having a small effect, to produce a trait. This results in continuous variation.
In epistasis, one allele masks or modifies the effect of another gene. This usually produces discontinuous variation (distinct categories of traits).

Coat Colour in Labrador

In many animals, coat colour is controlled by **epistasis**, where one gene affects another. For example, in Labrador dogs, coat colour (black, brown, yellow) depends on two genes.

1. The **B gene** controls pigment colour. Its dominant allele B gives black colour, while recessive b gives brown. So, BB or Bb will be black, and bb will be brown.
2. The **E gene** controls the deposition of pigment in fur. The dominant E allows pigment, but recessive e is epistatic to B or b. it blocks pigment deposition in fur. So, EE or Ee will pigment in fur, but ee will have yellow coat, no matter if B or b is present.

When two heterozygous black dogs (BbEe × BbEe) are crossed, the offspring show black, brown, and yellow colours in a 9:3:4 ratio.

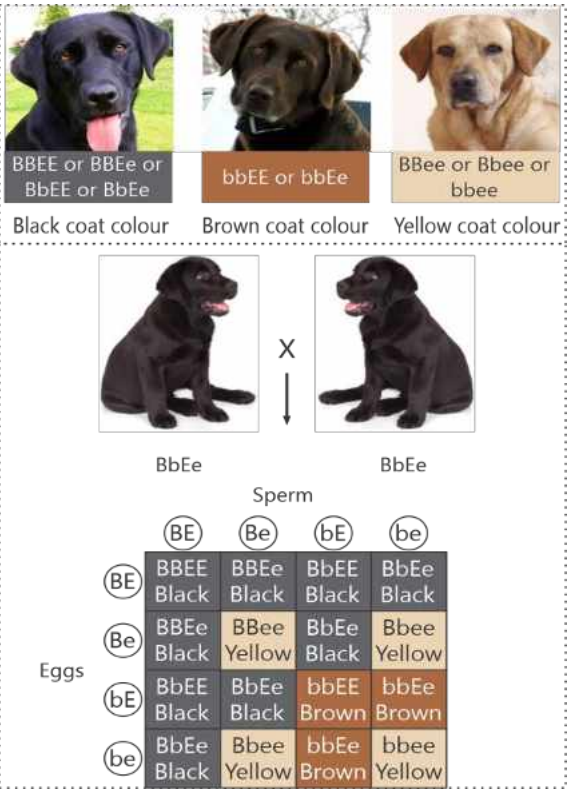


Figure 18.8: Genotypes and phenotypes of coat colour in Labrador

Bombay Phenotype

The expression of ABO genotypes also depends upon another gene i.e., H. It makes a particular H substance, which is a precursor to the A or B antigens. The alleles I^A and I^B produce enzymes which modify the H substance into antigen-A and antigen-B respectively. If an individual has I^A and I^B alleles but his genotype is hh, he will not produce H substance. So, his I^A and I^B alleles will not produce A and B antigens. In this way, he will have O phenotype due to the lack of A and B antigens. This phenotype is called Bombay phenotype because it was discovered in Bombay in 1952. It is mostly found in the India, Bangladesh, Pakistan, and Iran.

Flower Colour in Foxglove

In foxglove plants, the flower colour (dark red, light red, and white) is controlled by interaction in two genes (gene D and gene W).

1. Gene D has a dominant allele D that produces dark red petals. Its recessive allele d produces light red petals. Therefore, a genotype DD or Dd will have dark red and a genotype dd will have light red flower colour.
2. Gene W (present at another locus) has a dominant allele W that limits the distribution of pigment (produced by D and d) in the form of small spots so petals appear white from outside. The recessive w allows uniform pigment distribution.

The heterozygous plant DdWw will have white flowers (because the W allele limits the pigment in small spots in the inner side). If such heterozygous plant is self-fertilized, it produces plants with white, dark red and light red flowers in 12:3:1 ratio.

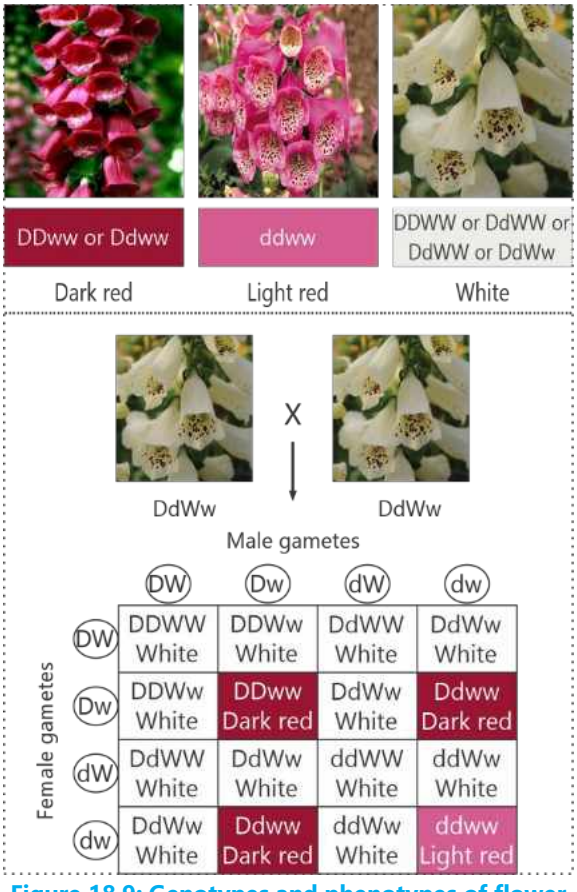


Figure 18.9: Genotypes and phenotypes of flower colour in foxglove

18.6- Gene Linkage and Crossing Over

In 1900, German geneticist **Karl Correns**, rediscovered Mendel's work and suggested the role of chromosomes in heredity. In 1902, American scientist **Walter Sutton** proposed the **chromosomal theory of inheritance**. This theory states:

- Genes are found at specific locations on chromosomes.
- The behaviour of chromosomes during meiosis explains Mendel's laws of inheritance.

18.6.1 Gene Linkage

In body cells, thousands of genes are present on a limited number of chromosomes. For example, humans have about 20,000 genes on 23 chromosome (present in two sets), and fruit fly (*Drosophila*) has about 13,000 genes on 4 chromosome (also present in two sets). This means many genes are arranged in a line on each chromosome, sometimes very close together. Genes present on the same chromosome are called **linked genes**, and this is known as gene linkage. Closely linked genes tend to stay together and do not assort independently during gamete

formation. However, genes on different chromosomes, or those far apart on the same chromosome, assort independently.

Detection of gene linkage

Gene linkage and its frequency can be studied by performing crosses and observing how often different phenotypes appear in the offspring. The key idea is to compare parental combinations (same as parents) with recombinant or new combinations.

For Information
Gene linkage can only be observed when a cross produces large number of offspring. Larger the progeny size, the more likely it is to represent the actual frequency of linkage in the population.

- If offspring show parental and new phenotypes in equal 1:1:1:1 ratio, it means the genes are not linked and assort independently.
- If parental combinations are more frequent and new combinations are fewer, it indicates incomplete linkage, meaning genes are linked but can separate due to crossing over.
- If only parental combinations appear and no new combinations are seen, it shows complete linkage, meaning the genes are very close and do not separate.

Example of Detection of Linkage

American biologist, **Thomas Hunt Morgan,** studied two traits in *Drosophila* i.e., wing length and abdomen width. He observed that allele for long wings (L) is dominant over short or vestigial wing (l). Similarly, allele for broad abdomen (B) is dominant over narrow abdomen (b). Morgan did the following cross in drosophila;

Parents		Broad abdomen Long wings		X		Narrow abdomen Vestigial wings	
				↓			
F1			Long wings Broad abdomens				

Then, he crossed two of F1 flies. The results are shown in the following figure:

Explanation of Result: According to law of independent assortment, the F2 generation should have the following 9:3:3:1 ratio:

F1 X F1	Long wings Broad abdomens	X	Long wings Broad abdomens
		↓	
F2	9 Long wings Broad abdomen (Parental combination)		1 Vestigial wing Narrow abdomen (Parental combination)
	3 Vestigial wings		3 Long-wings

	Broad abdomen (New combination)		Narrow abdomen (New combination)

But Morgan observed only parental combinations in F2. There were 3/4 flies with long wings-broad abdomen and 1/4 with vestigial wings-narrow abdomen.

F2	3 Long wings Broad abdomen (Parental combination)	1 Vestigial wing Narrow abdomen (Parental combination)

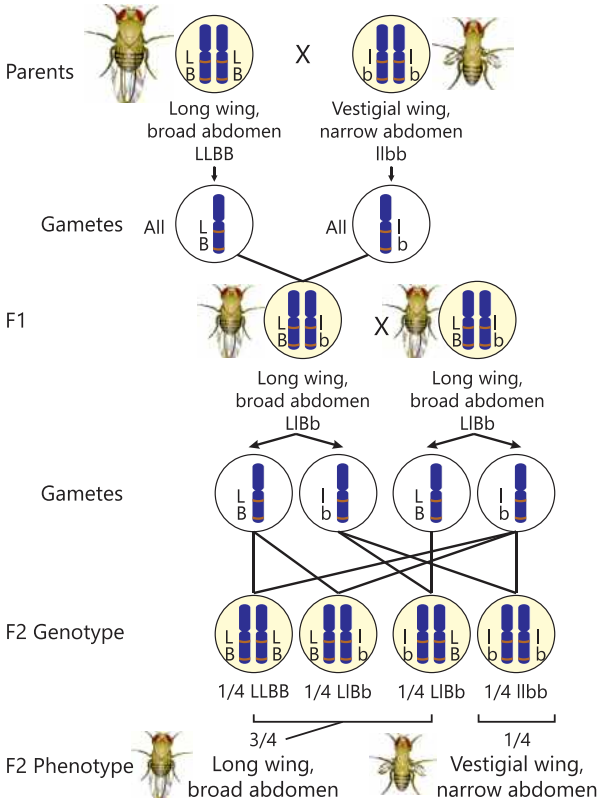


Figure 18.10: Gene linkage in Drosophila

From this data, Morgan concluded that the genes for wing length and abdomen width were located on the same chromosome and they did no assort independently.

18.6.2- Crossing Over

During the formation of gametes (meiosis-I), homologous chromosomes go through crossing over. This process is responsible for the recombination of linked

genes. During crossing over, the chromatids of two homologous chromosomes line up next to one another. They swap (exchange) sections of DNA. In this process, DNA strands actually break and re-join. After crossing over, the homologous chromosomes separate from each other. The chromatids involved in crossing over still have the same genes in the same order, but the alleles have been rearranged. When these recombinant chromatids are distributed in different gametes, a wide variety is produced in gametes. Therefore, crossing over leads to recombination of genes and thus responsible for genetic variability.

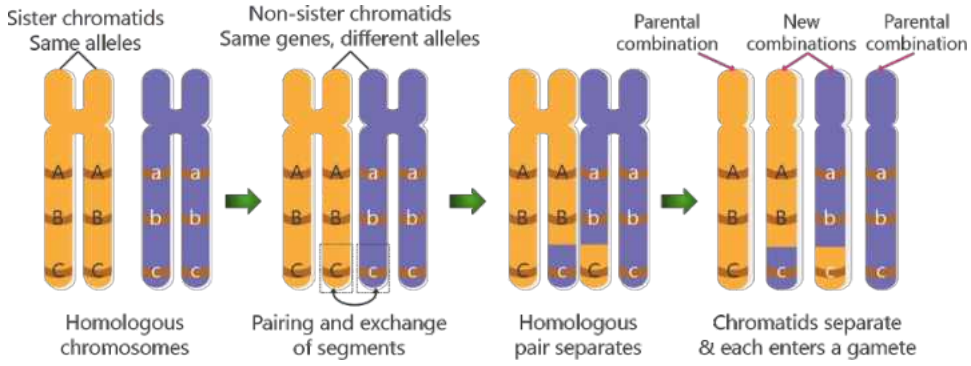


Figure 18.11: Crossing over

18.7- Sex Determination

The sex of an animal is determined during its embryonic development. In most animals, sex is determined on the basis of karyotype i.e., appearance of the complete set of chromosomes in the cells. Karyotype includes the numbers, sizes, and shapes of chromosomes. In the karyotype of most animals, all pairs of chromosomes are similar in female and male animals, except one pair. The chromosomes which are perfectly similar in female and male, are called **autosomes**. One pair of chromosomes is different in female and male and it determines the gender. The chromosomes of this pair are called **sex chromosomes**. The following are the main sex-determination systems on the basis of karyotype.

1- XX-XY System of Sex Determination

In *Drosophila*, mammals and most other vertebrates, the sex chromosomes pair consist of a rod-shaped **X chromosome** and a hook-shaped **Y chromosome**. Females have XX chromosomes while males have XY. Drosophila has 3 pairs of autosomes and 1 pair of sex chromosomes. Humans have 22 pairs of autosomes and 1 pair of sex chromosomes.

In these animals, females are **homogametic** which means that they produce all eggs with one X chromosome. On the other hand, males are **heterogametic** which means that they produce two types of sperm. Half carry one X chromosome and half

carry one Y chromosome. If a sperm with X chromosome fertilizes the egg, the zygote will have XX chromosomes. This embryo will develop female sex organs and the baby will be a female. If a sperm with Y chromosome fertilizes the egg, the zygote will have XY chromosomes. This embryo will develop male sex organs and the baby will be a male.

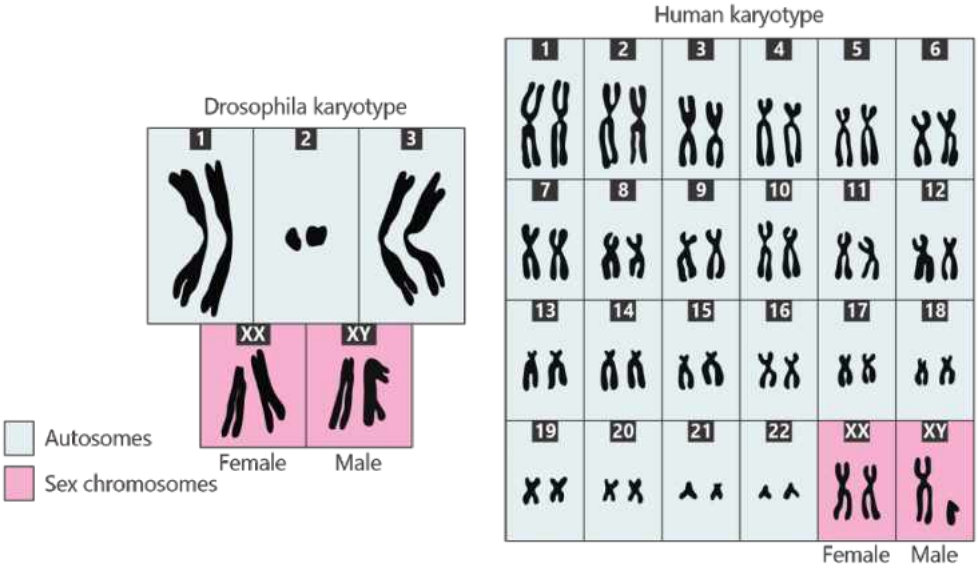


Figure 18.12: Autosomes and sex chromosomes in Drosophila and humans

2- XX–XO System of Sex Determination

This sex determination is found in some insects. For example, grasshopper has 22 autosomes (11 pairs). Male has total 23 chromosomes because its sex chromosome pair is XO i.e., only X chromosome is present and there is no Y chromosome. Male forms two types of sperm; half containing X and half without any sex chromosome. A gamete without any sex chromosome is called **nullo gamete**.

Female has 24 chromosomes. Its sex chromosome pair is XX. All eggs carry one X chromosome. If an X-carrying sperm fertilizes the egg, an XX female offspring is produced. If the nullo sperm fertilizes the egg, an XO male offspring is produced.

3- ZW–ZZ System of Sex Determination

This sex determination is common in birds. The autosome number differs in different birds but the sex chromosomes are ZW in females and ZZ in males. It means that the female bird produces two kinds of eggs; half with Z chromosome and half with W chromosome. While, the all sperms of the male have Z chromosome. When an Z carrying egg is fertilized by the sperm, offspring is male. When a Y carrying egg is fertilized by the sperm, offspring is female.

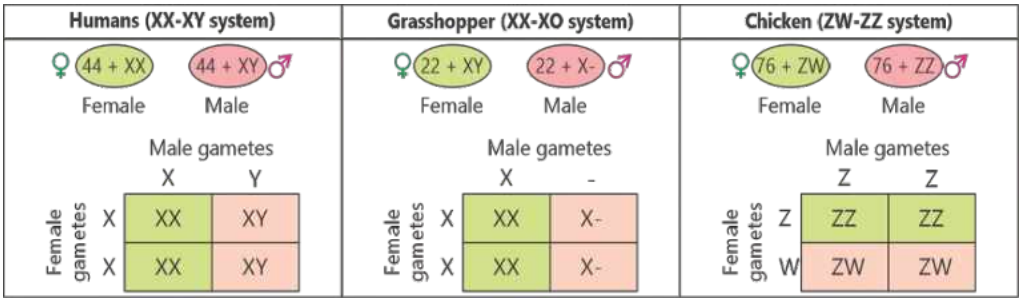


Figure 18.13: Sex determination in animals

18.8- Sex Linkage

The traits which are controlled by alleles present on sex chromosomes are called **sex-linked traits** and the phenomenon is called sex linkage. The sex-liked trait is actually **X-linked** because usually the Y chromosome carries almost no functional genes. That's why, the ratio of sex-linked traits is different in males and females. However, the alleles of very few traits are found on both X and Y. Such traits are equally distributed among males and females and are called **X-Y-linked** or **pseudo-autosomal** traits.

Discovery of Sex-Linkage

In 1910, Thomas Hunt Morgan observed a male *Drosophila* fly that differed from normal flies. This mutant male fly had white eyes instead of normal red. He crossed the mutant male to a normal (wild-type) female. All of the F1 progeny had red eyes.

Parents	1 White-eyed (male)	n	X	1 Red-eyed (female)
		↓		
F1	All Red eyed (male)	All Red-eyed (female)		

He concluded that red eye colour was dominant over white. He crossed the red-eyed flies from the F1 generation with each other. Out of 4252 flies in F2, there were 782 (18%) white eyed. Although the ratio of red eyes to white eyes in the F2 was greater than 3:1, there was something important. All of the white-eyed F2 flies were males.

F1 X F1	1 Red-eyed (male)	X	1 Red-eyed (female)
		↓	
F2	3473 Red eyed (male & female)	782 White-eyed (male)	

Morgan thought that perhaps it was impossible for a white-eyed female fly to exist. To test this idea, he test-crossed the F1 red-eyed female with the white-eyed male. He obtained both white-eyed and red-eyed males and females in a 1:1:1:1 ratio. Hence, it was proved that a female could have white eyes. Why, then, were there no white-eyed females among the progeny of the first cross? He mated a white-eyed female with a red-eyed male. All female offspring had red eyes, and all male offspring had white eyes.

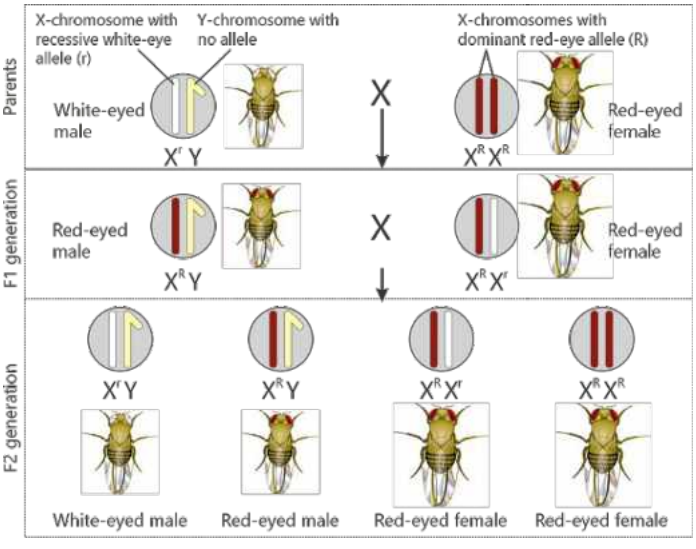


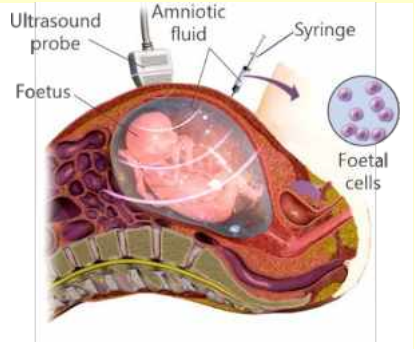
Figure 18.14: Inheritance of sex-linked trait (eye colour) in Drosophila

Morgan concluded that the gene of white-eye is present on X chromosome. He assumed that the Y chromosome did not have the allele of this gene. He called such genes as sex-linked genes and traits as sex-linked traits.

Embryonic Screening

Embryonic screening are the methods to check genetic disorders and abnormal number of chromosomes in embryos. It includes methods like ultrasound scan, chorionic villus sampling, and amniocentesis.

Amniocentesis is generally done during 15th and 20th week of pregnancy. In its procedure, a long needle is inserted into the amniotic sac. During it, an ultrasound probe locates the foetus and tests any abnormality. The inserted needle sucks some amniotic fluid which contains cells of the foetus. The foetal cells are kept in culture medium and checked for abnormal number of chromosomes.



Amniocentesis

18.8.1- Sex–linked Traits in Humans

X-linked Recessive Traits

The X-linked recessive traits are less common in females than males. It is because, such trait will appear in a female only when she has two recessive alleles on her two X

chromosomes. While in males, this X-linked recessive trait will appear when only one allele is present on X-chromosome (because the Y-chromosome does not carry its allele).

So, this trait will appear in all the males who possess its allele. Examples of X-linked recessive traits include muscular dystrophy (muscle weakness and degeneration), red-green colour blindness and haemophilia (deficiency of clotting factors in the blood).

If a mother has the trait, all of her sons will also have it. If mother is a carrier (heterozygous) of the trait, 50% of her sons can inherit it. If such trait is present in father, all of his daughters will be carriers (daughters possess their father's X-chromosome) and no son will inherit this trait (sons do not inherit their father's X-chromosome).

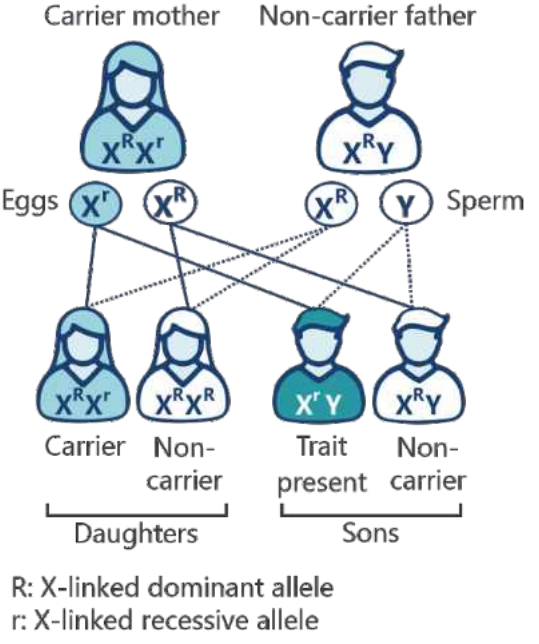


Figure 18.15: Inheritance of X-linked recessive traits

X-linked Dominant Traits

Such traits are in equal ratio in females and males. Examples include vitamin D-resistant rickets (inability to properly metabolize vitamin D, leading to softening and weakening of bones), Rett syndrome (neuro-developmental disorder with loss of speech, repetitive hand movements, and problems with walking), Aicardi syndrome (abnormal development of brain).

If an X-linked dominant trait is present in mother (homozygous or heterozygous), 50% of her offspring can inherit the trait. If such trait is present in father, all of his daughters will inherit it (a daughter inherits her father's X-chromosome) and no son will inherit it (a

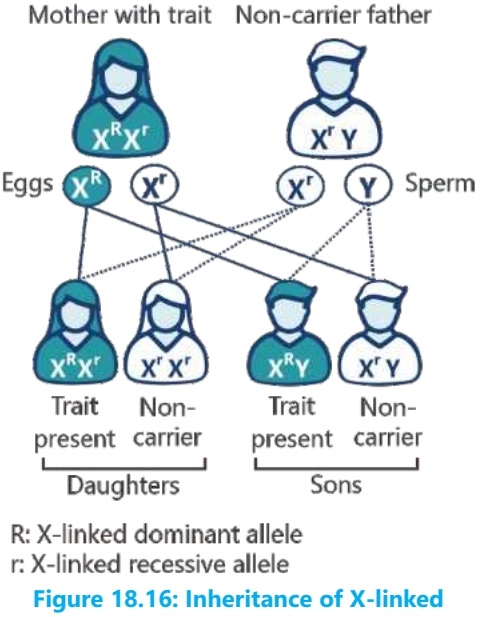


Figure 18.16: Inheritance of X-linked dominant traits

son does not inherit his father's X-chromosome).

Y-linked Traits

Y-chromosome contains relatively few genes compared to X chromosome. That's why Y-linked traits are relatively rare. The Y-linked genes do not have alleles on X chromosome. Such genes are called holandric genes (or hemizygous genes). Such genes are located only on the non-homologous region of Y-chromosome. The Y-linked traits appear only in males and are inherited from father to son only. The examples of Y-linked traits in human are hypertrichosis (growth of hair on the rim of pinna), porcupine man (straight hair on the body) and webbing of toes.

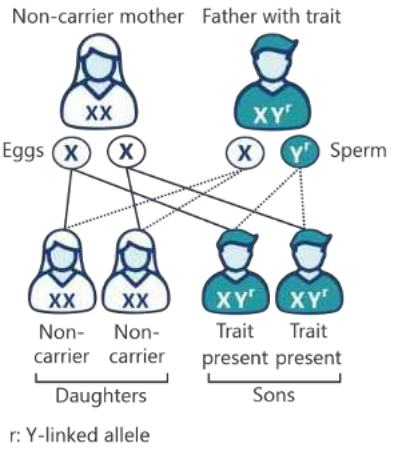


Figure 18.17: Inheritance of Y-linked traits

18.8.2- Sex Linked Disorders in Humans

1- Haemophilia

Haemophilia is a rare X–linked recessive trait. In this disease, the blood fails to clot properly after an injury. It is due to the reduction, malfunctioning, or absence of blood clotting factors. Haemophilia is of three types i.e., A, B and C.

Haemophilia A and B are due to abnormal blood clotting factors VIII and IX, respectively. Both these types are X–linked recessive traits. So, these are more common in males than females.

Haemophilia C is due to abnormal blood clotting factor XI. It is an autosomal recessive trait, so present in both genders equally.

For Information

In human males, there is a specific gene on Y-chromosome. It is known as SRY (Sex-determining Region Y). It produces a protein which starts development of testes and prevents the development of female reproductive structures.

Solve the following genetic problem.

If two normal parents have a haemophiliac child, what could be the genotypes of the parents?

Mechanism of Normal Blood Clotting (Coagulation)

Blood clotting is a complex process that helps to prevent excessive bleeding when an injury occurs. The mechanism involves the following steps:

1. When an injury occurs, platelets adhere to the site of injury, forming a platelet plug.
2. Activated platelets as well as the damaged tissue release of clotting factors.
3. The clotting factors perform chemical reactions (clotting cascade), in which soluble protein fibrinogen is converted into an insoluble protein called fibrin.

4. Fibrin forms a mesh-like structure that entraps parts of the blood and makes a solid clot.
5. The clot is stabilized by the cross-linking of fibrin strands.

If the balance of clotting system is disrupted, it can lead to abnormal clotting, known as **thrombosis**, which can cause serious health problems such as stroke and heart attack.

2- Colour-Blindness

In people with normal vision (**trichromatic** vision), there are three kinds of cone cells in the retina of eye – each sensitive for a specific primary colour. These are red-sensitive cones, green-sensitive cones and blue-sensitive cones. Each type contains its specific pigment proteins called **opsins**. The genes for red and green opsins are on X chromosome, while the gene for blue opsin is on autosome 7. The recessive alleles for opsins cause colour-blindness. It may be of following types.

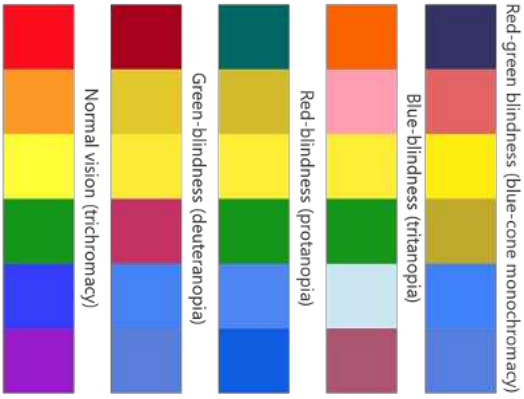


Figure 18.18: Dichromacy & monochromacy

Dichromacy: It is the perception of two primary colours and blindness of the third colour whose opsin is missing. It may be red-blindness (protanopia), green blindness (deuteranopia), or blue-blindness (tritanopia).

Monochromacy (total colour blindness)**:** It is the perception of a single colour and blindness of any two colours whose opsins are missing. In blue-cone monochromacy, red and green opsins are absent. It is also called red–green colour-blindness. Red-cone monochromacy and green-cone monochromacy are very rare.

There is no cure for colour blindness. However, there are assistive technologies such as colour-blind glasses and special software that can help individuals to differentiate between colours. Additionally, some colour-blind people have found success with colour discrimination training.

3- Muscular Dystrophy

There are more than 30 disorders classified as muscular dystrophies. **Duchenne muscular dystrophy** is the most common. It is X-linked recessive (so more common in males). About two thirds cases are inherited from a person's mother, while one third cases are due to a new mutation. If a male receives its recessive allele, he cannot make muscle proteins (dystrophin). It results in the death of muscle fibres and there is weakness and breakdown of skeletal muscles, especially those of the hips, pelvic area, thighs, and calves. It eventually progresses to the shoulders and neck, arms, respiratory and cardiac muscles, and other areas.

Muscular dystrophy affects a child around the age of four. It becomes evident when the child begins walking. By age 10, the child may need braces. By age 12, most patients are unable to walk and need wheelchair. Typical lifespans range from 15 to 45.

There is no cure for muscular dystrophy, but treatments are available to manage its symptoms and improve quality of life. For example; physical therapy is done to maintain strength, flexibility, and mobility; corticosteroid medicines can give short-term improvements in muscle strength and function; pain-killer medicines are used to relieve muscle pain; and surgery is done to correct joint contractures.

Muscle weakness | Abnormal calf muscles | Fatigue | Walking on toes | Breathing problems

Figure 18.19: Symptoms of muscular dystrophy

18.8.3- Sex-limited and Sex-influenced Traits

Some traits are not sex-linked (because their alleles are not present on sex chromosomes) but there ratio is different in males and females. There are two categories of such traits.

1- Sex-limited Traits

The genes of such traits are present in both sexes but are expressed in only one sex due to anatomical or hormonal differences. Such traits are called sex-limited traits. For example, both male and female sheep have the genes of horn development. These genes are only expressed in male sheep. Similarly, the genes of milk production are present in both male and female mammals. But, these genes express only in female mammals. The genes of the growth of beard in humans are expressed in males only. Similarly, the genes of prostate cancer are expressed in males only and genes of ovarian cancer are expressed in females only, although both these genes are present in males and females.

For Information

Sex-limited genes are responsible for sexual dimorphism i.e., phenotypic difference between males and females of the same species regardless of genotype.

2- Sex-influenced Traits

In some traits, the same gene shows different expression in each sex. Sex-influenced genes have alleles which are dominant in one sex and recessive in the other. This difference in expression is due to hormonal difference between the males and females. For example, in humans, the traits of the amounts of body hair and baldness are sex-influenced traits. The allele of baldness is dominant in men but recessive in women. So, a man can be bald when he is homozygous (BB) or heterozygous (Bb) for baldness alleles. While a woman can be bald only when she inherits two recessive alleles (bb).

For Information

Genetics has made significant progress. Now, it is more applied and practical science. For example:

- DNA sequencing and genetic engineering, have enabled researchers to manipulate genes.
- The completion of Human Genome Project has led to a deeper understanding of the genetic basis of various diseases and traits.
- Genetics plays important role in healthcare, helping doctors to diagnose and treat patients based on their unique genetic profiles.

EXERCISE

SECTION 1: MULTIPLE CHOICE QUESTIONS

1. In the monohybrid cross of Mendel, when a pure round-seeded plant was crossed with a winkled-seeded plant, all the F1 progeny were round-seeded, because of:
   (a) Co-dominance (b) Incomplete dominance
   (c) Complete dominance (d) Epistasis
2. In a cross of AaBb x AaBb, what fraction of the offspring can be expected to express one of the two dominant alleles, but not both?
   (a) 1/16 (b) 6/16 (c) 9/1 (d) 16/16
3. In which of these cases, two genes can experience independent assortment?
   (a) They are located in close proximity on the same chromosome.
   (b) Crossing over between the genes does not occur.
   (c) They are located on different chromosomes or are far apart on the same chromosome.
   (d) The expression of one gene does not affect the expression of the other.
4. The blood group AB may be given to a person who has blood group;
   (a) A (b) O (c) B (d) AB
5. If father has blood group B and the mother has blood group A, what blood groups their children have?
   (a) A or B only (b) AB only (c) AB or O only (d) A, B, AB or O

6. A gene for corn has two alleles, one for yellow kernels and one for white kernels. Cross pollination of yellow corn and white corn results in ears with approximately equal number of yellow and white kernels. Which term best describes the relationship between the two alleles?
   (a) Crossing over (b) Recombination
   (c) Incomplete dominance (d) Codominance
7. A cross between two plants with genotypes AaBb and AaBb results in an F1 generation that is 25% AABB, 50% AaBb, and 25% aabb. Which reason most likely explains why other possible genotypes are not present?
   (a) The genes underwent independent assortment
   (b) The loci of the genes are close together.
   (c) The loci of the genes are on different chromosomes.
   (d) Crossing over occurred between chromosomes.
8. In pea plants, the allele for tall stalks (T) is dominant over the allele for short stalks (t). Suppose a cross between a tall pea plant and a short pea plant produces 43 tall offspring and 47 short offspring. If one of the tall offspring is crossed with one of the short offspring, what ratio of genotypes would be most likely in their offspring?
   (a) 1 TT : 2 Tt : 1 tt (b) 3 Tt : 1 tt
   (c) All Tt (d) 1 Tt : 1 tt
9. Allele B produces black coat colour while b produces white coat colour. Allele C produces curly fur while c produces straight fur. In a cross between two BbCc parents, predict the fraction of offspring with black coat colour and straight fur.
   (a) 1/16 (b) 9/16 (c) 3/4 (d) 16/16
10. What is the criterion to mark an allele as dominant?
   (a) The frequency of the allele in population
   (b) The expression of the allele in heterozygous genotype
   (c) The location of the allele on chromosome
   (d) The frequency of the crossing over of the allele
11. Which of these characters is controlled by a multiple allelic system?
   (a) ABO blood group system (b) Rh blood group system
   (c) MNS blood group system (d) All of these
12. Albinism is caused by a recessive gene. A woman with albino father marries an albino man. What will be the proportion of her progeny?
   (a) 2 normal : 1 albino (b) All normal
   (c) All albino (d) 1 normal : 1 albino
13. When the activity of one gene is suppressed by the activity of a non-allelic gene, it is known as;
   (a) Incomplete dominance (b) Epistasis
   (c) Complete dominance (d) Multiple allelic effect

14. Which of the following is a result of Erythroblastosis fetalis?
    (a) Anaemia in the foetus (b) Jaundice in the foetus
    (c) Mental retardation in the foetus (d) All of these
15. Colour blindness is more common in males because it is a/an;
    (a) Autosomal trait (b) X-linked dominant trait
    (c) X-linked recessive trait (d) Y-linked trait

SECTION 2: SHORT QUESTIONS

1. Name the main exceptions to the Mendel's laws of inheritance.
2. What is probability? How does probability relate to genetics?
3. Justify that multiple alleles provide many different phenotypes.
4. Name various human blood group systems.
5. What do you mean by multiple alleles? Name the alleles responsible for ABO blood group system.
6. What is co-dominance. Give an example.
7. What antigens and antibodies are found in ABO blood group system?
8. Justify why O-negative individuals are called universal donors and AB-positive individuals are called universal recipients.
9. Why is human male referred as heterogametic?
10. What is gene linkage?
11. How can the linked gene assort independently?
12. List at least five polygenic traits in humans.
13. Write brief note on Y-linked inheritance in humans.
14. Define sex linkage and give an example.
15. Solve the following genetic problems:
    i. A plant of genotype AABbCC is self-pollinated. Calculate the phenotypic ratio of F2 generation.
    ii. In four-o'clock plant, the red and white flower colours show incomplete dominance. Calculate the ratios in crosses between (i) two red-flowered plants; (ii) a red-flowered and a pink-flowered plant; (iii) a pink-flowered and a white-flowered.
    iii. A man with blood group A marries a woman who has blood group AB. What blood groups are expected among their children?
    iv. Two white sheep produce a black offspring. What are the parents' genotypes for the colour? What is the probability that their next offspring will be black?
    v. Albinism (lack of pigment) in man is caused by a recessive gene. If normal parents have an albino child, what is the probability that their next child will be normal for colour?
16. Differentiate between:
    - Gene and allele
    - Genotype and phenotype

- Homozygous and heterozygous
- Incomplete dominance and co-dominance
- XX-XY and ZZ-ZW sex determination
- Sex-linked and sex-limited traits
- Monohybrid and dihybrid cross
- Epistasis and polygenic inheritance
- XX-XY and XX-ZO sex determination
- Sex-limited and sex-influenced traits

SECTION 3: LONG QUESTIONS

1. Explain Mendel's experiment which led to the formulation of the law independent assortment.
2. Explain incomplete dominance and exemplify it through the inheritance of flower colour in 4 O' clock plant.
3. Explain Erythroblastosis foetalis in the light of antigen-antibody reaction. What measures can be taken to prevent Erythroblastosis foetalis?
4. Describe polygenic inheritance, using an example from grain colour in wheat.
5. Explain epistasis with the help of an example.
6. Explain how gene linkage counters independent assortment and crossing-over modifies the progeny.
7. Exemplify the concept of gene linkage by quoting the example of wing length and width of abdomen in Drosophila melanogaster.
8. Explain the XX-XY mechanism of sex determination in Drosophila and mammals.
9. Describe the XX-XO and ZZ-ZW sex determination systems and evaluate by studying the karyotype.
10. Explain the inheritance of a sex-linked trait in Drosophila.
11. Describe sex-influenced and sex-limited traits with common examples from human genetics.
12. Write a note on the inheritance of haemophilia in humans.
13. Write a note on the inheritance of colour blindness in humans.
14. Explain Morgan's experiment to explain crossing over and gene linkage.

INQUISITIVE QUESTIONS

1. Collect data from the class or the college to see how many individuals have AB blood group and construct a pie chart and histogram for the collected data.
2. Use a dice to calculate how many times out of 100 throws can you get sixes.
3. Derive an idea to get alternatives of blood transfusion. (reference could be made to synthesized plasma and serum).
4. Justify why a recessive blood group allele of 'i' is more frequent in population.
5. Justify blood donation as a service to suffering humanity.
6. Suggest ways to save lives through the knowledge gained in this chapter.

Chapter 19 CHROMOSOME AND DNA

STUDENTS'LEARNING OUTCOMES

After studying this chapter, the students will be able to:

- Annotate the detailed structure of a chromosome.
- Narrate the experimental work of Griffith and Hershey-Chase, which proved that DNA is the hereditary material.
- Describe the three models proposed about the mechanism of DNA replication.
- Describe the events of the process of DNA replication.
- Describe DNA stability and variability as two characters of the replicating DNA molecule.
- Describe the characteristics of genetic code (universal, triplet, non-overlapping, degenerate, has no punctuation).
- Differentiate between the terms genetic code and codon.
- Explain the mechanism of transcription.
- Explain why the length of transcribed mRNA molecule (in Eukaryotes) shortens as it enters the cytoplasm for translation.
- Describe the mechanism of protein synthesis.
- State the difference between protein synthesis in prokaryotes and eukaryotes.
- Suggest possible ways in which the synthesized protein can be used within or outside a cell that synthesized it.
- State the importance of the regulation of gene expression.
- Describe the negative control of gene expression by repressor proteins.
- Describe the positive control of gene expression by activator proteins.
- Define mutation and identify various sources of mutation.
- Differentiate between natural and induced mutations and mutagens.
- Justify most mutations are harmful.
- Rationalize that mutations might be a contributing factor towards evolution.
- Describe the symptoms, causes and possible available treatments of some of the
- chromosomal mutations. (Down's, Klinefelter's and Turner's syndrome).
- Describe the symptoms, causes. and possible available treatments of some of the gene mutations.

You have the basic knowledge of chromosomes and DNA. You know that genes are specific segments of DNA which carry information for the making of a protein. This chapter aims the detailed study of genes. We will also study the metabolic processes involved in gene working.

19.1- Detailed Structure of Chromosome

The structure and chemical composition of chromosomes are different in prokaryotic and eukaryotic cells. In this section, we will study chromosomes in eukaryotic cells.

Tidbit
Chromosomes were discovered by a German embryologist, **Walther Flemming** in 1882. He discovered chromosomes in the rapidly dividing cells of salamander larvae. The name "chromosome" was proposed by German anatomist, **Waldeyer-Hartz** in 1888. "Chromosome" literally means coloured bodies.

Chromosomes are thin, thread-like structures. They are made of DNA and histone proteins. The number and shape of chromosomes are different in each species. This complete set is called the **karyotype**. In body cells (somatic cells) of diploid organisms, chromosomes occur in pairs. These pairs are called homologous chromosomes. Gametes (sex cells) have half the number of chromosomes. These chromosomes are not in pairs.

When a cell is not dividing (interphase), its chromosomes are not visible as distinct structures. They remain uncoiled and form a fine network, called **chromatin**. When a cell prepares to divide, the DNA starts to coil. It wraps around small units of histones. These units then coil further into highly condensed form. With more coiling and condensation, the structure becomes thicker and shorter. Finally, it forms compact thread-like structures i.e., chromosomes.

19.1.1- Morphology of Chromosomes

During interphase, the duplication of chromatin also occurs. It involves the replication of DNA and synthesis of new histone proteins. After duplication, each chromosome consists of two identical strands called **chromatids**. These two strands are known as sister chromatids. They are attached to each other at a constricted region called the **centromere**. The centromere is a specific DNA sequence of about 220 nucleotides. A disc-shaped protein structure, called the **kinetochore**, is attached to the centromere. It serves as the point where chromosomes attach to the mitotic spindle during cell division.

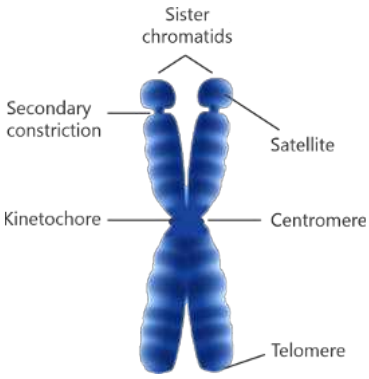


Figure 19.1: Morphology of a duplicated and condensed chromosome

In some chromosomes, there may be another constriction, called **secondary constriction** or **Nuclear Organizer Region (NOR)**. It makes nucleoli during interphase. The end of chromosome beside secondary constriction is knob-like. It is called **satellite**. This

region has highly repetitive DNA sequences called junk DNA. The terminal ends of chromosomes are called **telomeres**. They prevent the two chromosomes to attach with each other from their ends.

The position of centromere is different in different chromosomes. The chromosomes in which centromere is at the centre are called **metacentric** chromosomes. The ones in which centromere is located slightly away from centre are **sub-metacentric** chromosomes. In **acrocentric** chromosomes, centromere is located near the end, and in **telocentric** chromosomes, centromere is located at the end.

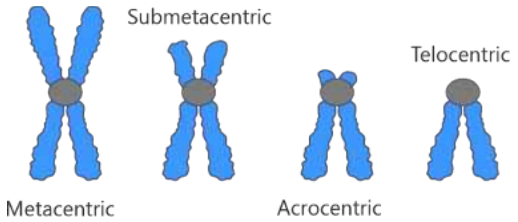


Figure 19.2: Types of chromosomes on the basis of location of centromere

19.1.2- Chemical Composition and Organization of Chromosome

A chromosome is made up of **40% DNA** and **60% proteins (histones)**. Both chromatids have identical DNA molecules. The DNA in an average human chromosome is about 5 cm long and consists of about 140 million nucleotides. DNA is a negatively charged due to the presence of phosphate groups. It has strong affinity to **histone** proteins, which are positively charged due to the abundance of basic amino acids such as arginine and lysine. The following are the levels of the organization of chromosomes.

1- Nucleosome String

After replication, approximately every 146 nucleotides of DNA (2 nm thick) wrap twice around a histone octamer (set of 8 histone molecules i.e., two copies each of the histones H2A, H2B, H3, and H4). So, aggregations called **nucleosomes** are formed. Nucleosomes are connected by about 80 nucleotide long **linker DNA**. The fifth type of histone i.e., H1 called **linker histone** is present at the base of the nucleosome near the DNA entry and exit points. It helps in the winding of the chain of nucleosomes. All nucleosomes (without linker histone) resemble beads on a string called nucleosome string (11 nm thick).

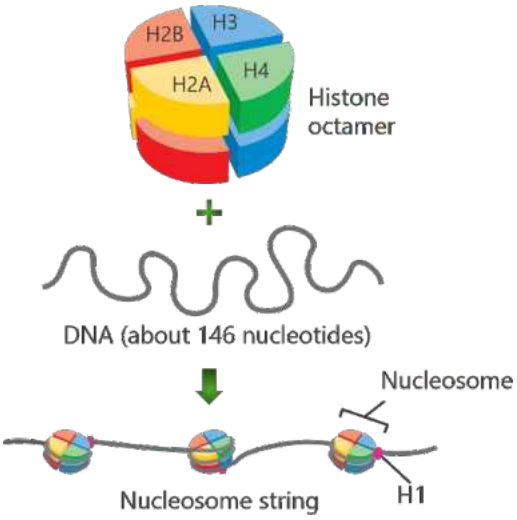


Figure 19.3: Nucleosome string

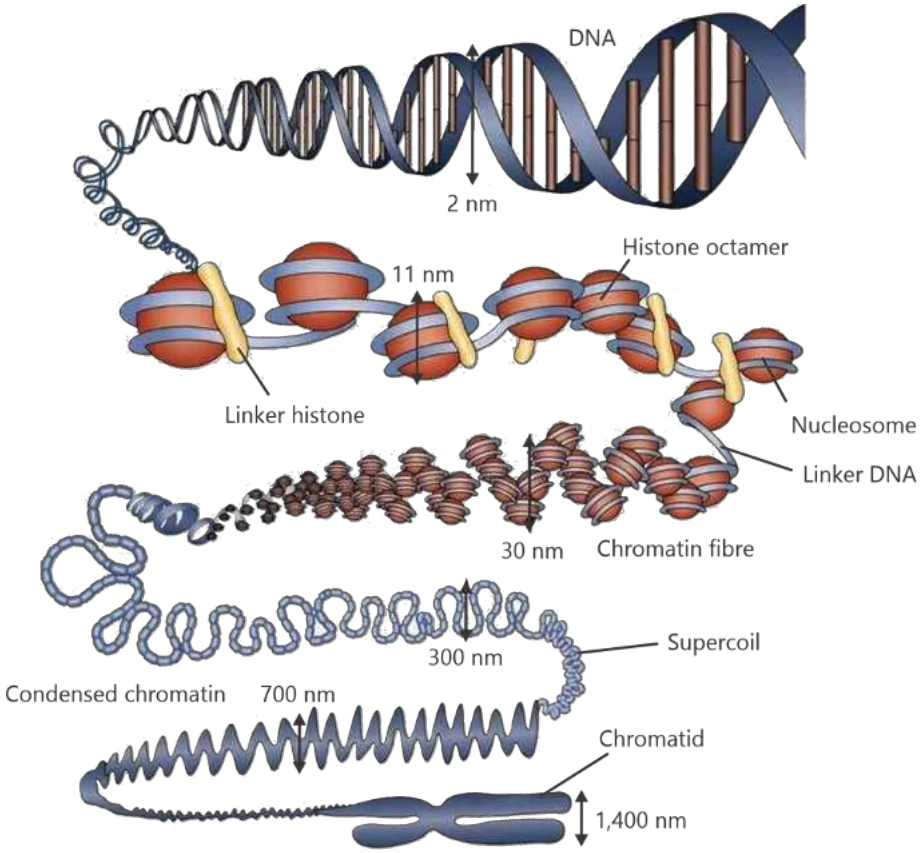


Figure 19.4: Organization of chromosome

2- Chromatin Fibre

In the early stages of cell division, the nucleosome string again coils about its axis. Tightly packed (condensed) nucleosomes make a thicker fibre called chromatin fibre (30 nm).

Tidbit

There are two regions in chromatin. The inactive portions of chromatin produce dark bands and are called **heterochromatin**. Whereas active portions of chromosomes are called **euchromatin**.

3- Chromatids

The chromatin fibre folds upon itself to make a supercoil (300 nm thick). The supercoil further coils and condenses and takes the shape of chromatid (700 nm thick).

19.1.3- The Concept of Genes and Alleles

In 1909, a Danish botanist **Wilhelm Johannsen** introduced the term "gene" for the basic unit of heredity. It was found that a gene is “a short segment of DNA which encodes the sequence of amino acid of a particular polypeptide”. The whole DNA molecule present in a chromosome does not contain message for sequencing amino

acids (preparing proteins). Rather, short segments of DNA, called genes, encode proteins. The locations where a gene is present on chromosome is called its **locus** (plural: loci).

On a homologous chromosome the alternate forms of a gene i.e., **alleles** are present on the same locus. For example, if a chromosome has the allele for 'eye colour' at a particular locus, its homologous chromosome will carry the allele of same character at the same locus.

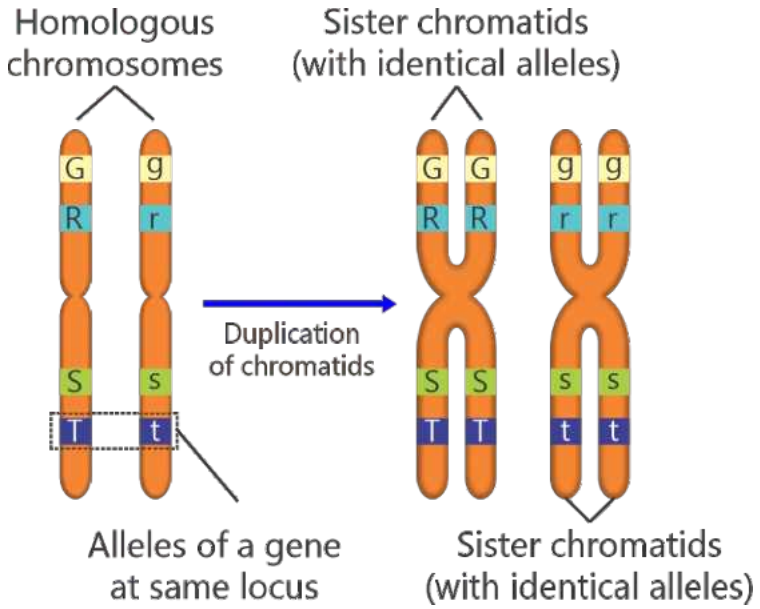


Figure 19.5: Alleles on homologous chromosomes

19.2- DNA AS THE HEREDITARY MATERIAL

After the confirmation of the following facts;

- Genes are the units of inheritance,
- Genes are located on chromosomes, and
- Chromosomes are made of DNA and proteins,

scientists tried to determine whether genes are made of DNA, or protein or both. It took almost 30 years (1920s – 1950s) to solve this question.

Information

A molecule that serves as genetic material must have certain characteristics. For example:

- It must be able to code for the sequence of amino acids in proteins.
- It must be able to replicate itself prior to cell division.
- It must be in the nucleus of eukaryotic cells.
- It must be able to change over time to account for evolution.

Only one molecule i.e., DNA fulfils all these requirements.

19.2.1- Griffith's Experiment

In 1928, British biologist, **Fredrick Griffith**, worked on a pathogenic bacterium *Streptococcus pneumoniae*. He observed two strains of this bacterium;

"S-type" strain had a polysaccharide coat and its colony appears "smooth".

"R-type" strain lacked polysaccharide coat and forms a "rough" colony.

When Griffith injected S-type bacteria into mice, they proved virulent (lethal). The mice developed pneumonia and died. But, when he injected R-type bacteria in mice, the mice showed no effects. Griffith thought that the virulent effect of S-type might be due to its polysaccharide coat which protects it from host's immune system. Then, he injected heat-killed S-type bacteria into mice, they remained healthy. To further disclose the reasons of virulence, he injected a mixture of heat-killed S-type (virulent, but killed) and alive R-type (non-virulent) into mice. He expected that the mice should not develop the disease. But the mice developed pneumonia and died. When he observed the blood of these dead mice, he found many alive S-type bacteria in the blood of dead mice.

Griffith concluded that the information for making polysaccharide coat and virulence had passed from the heat-killed S-type bacteria to the alive R-type. So, R-type bacteria made polysaccharide coat and transformed into virulent S-type. He called this process as **transformation** i.e., the transfer of genetic material from one organism to another to change the genetic makeup of the recipient organism. Griffith could not find the chemical nature (DNA or protein or both) of the material that was transferred from dead S-type to the alive R-type bacteria.

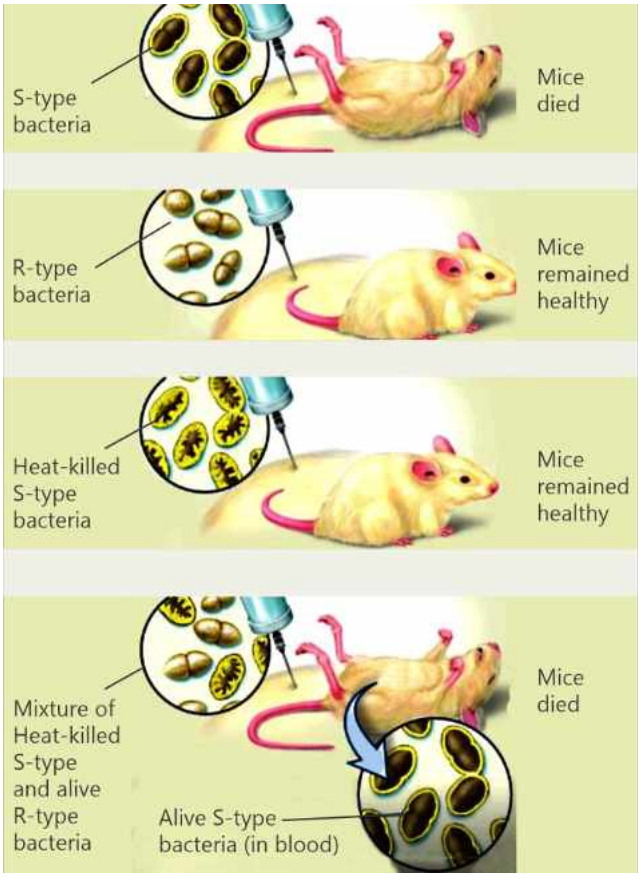


Figure 19.6: Griffith's experiment

Information

In 1944, three scientists, Avery, MacLeod and McCarty discovered the transforming principle. They removed proteins from the dead S-types bacteria and mixed them with alive R-type. When they injected this mixture into mice, they developed disease and alive S-type bacteria were found in them. It means that proteins are not responsible for transforming R-type into S-type.

Then, they removed all DNAs from dead S-type. They mixed S-type (without DNA) with alive R-type and injected into mice. They observed that no transformation had occurred this time as the mice remained healthy.

They concluded that "DNA is the fundamental unit of the transforming principle".

19.2.2- Hershey-Chase Experiments

Alfred Hershey & Marha Chase

In 1952, two American biologists, Alfred **Hershey** and Martha **Chase**, conducted experiments to confirm that DNA is genetic material. Their main experiment was with bacteriophages (viruses that attack bacteria).

Background Information: Bacteriophages consist of a core (made of DNA or RNA) and a protein coat. When a lytic bacteriophage infects a bacterium, it binds to the bacterial surface and then injects some material into the cell. The material is hereditary in nature because it directs the production of new bacteriophages within the bacterium. The bacterial cell eventually ruptures, or lyses and new bacteriophages are released.

Question: Which material (DNA/RNA, protein, or both) bacteriophages inject into bacterial cells that produce new bacteriophages?

Experiments: Hershey and Chase used a bacteriophage that contained DNA rather than RNA. They labelled the DNA and protein of bacteriophage with different radioactive isotopes. The isotopes would serve as **Tracers**. For this purpose.

- Some bacteriophages were grown on a medium containing an isotope of sulphur (^{35}S). The isotope was incorporated into the amino acids of the protein coats of new bacteriophages.
- Some bacteriophages were grown on a medium containing an isotope of phosphorus (^{32}P). The isotope was incorporated into the phosphate groups of the DNA of new viruses.

They prepared 2 setups.

1. In setup 1, they allowed bacteriophages with ^{35}S labelled protein to infect bacteria.

2. In setup 2, they allowed bacteriophages with ^{32}P labelled DNA to infect bacteria.

After some time, the infected bacterial cells and medium of both setups were separated by using centrifugation technique. The bacterial cells and media were analysed for the presence of radioactivity.

- In setup 1 (with bacteriophages containing ^{35}S labelled protein), they found radioactivity in the medium and not in the bacterial cells.
- In setup 2 (with bacteriophages containing ^{32}P labelled DNA), radioactivity was observed in bacterial cells and not in the medium.

These observations showed that during infection, ^{32}P labelled DNA of bacteriophages entered into the bacterial cells and directed the making of new bacteriophages. While, ^{35}S labelled protein coat remained outside.

Based on these observations, Hershey and Chase claimed that the DNA of bacteriophages, not the protein, was responsible for directing the production of new bacteriophages.

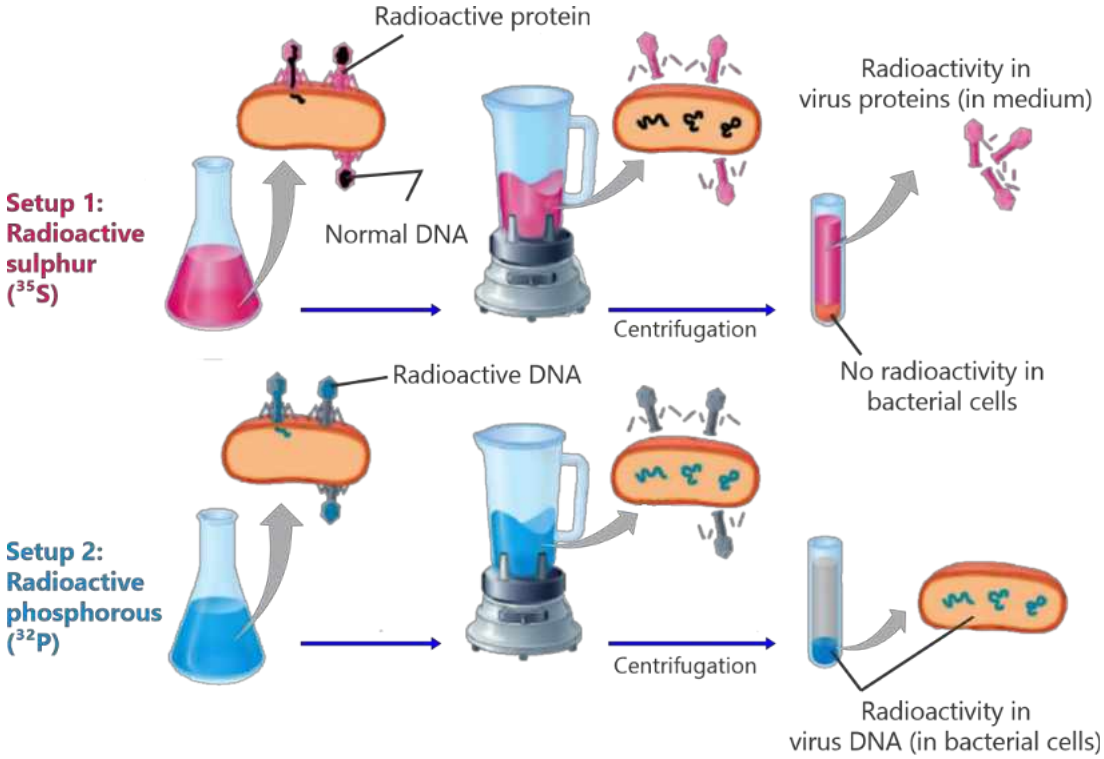


Figure 19.7: Hershey-Chase experiment

19.3- DNA Replication

DNA replication is the process in which the molecule of DNA makes its identical copy (replica). DNA is a double-helix molecule (made of two strands). So, it makes copies of both of its strands.

Recalling:
The DNA present in chromatids replicates so that chromatids can duplicate. It happens before a cell divides i.e., during the S-phase of interphase.

19.3.1- Models Proposed for the Mechanism of DNA Replication

Three models were proposed for the basic mechanism of DNA replication.

1- Conservative model: According to this model, the double-helix of parent DNA remains intact. This double-helix generates new double-helix which completely consists of new material. So, after replication, when two daughter DNAs are produced;

- 1 daughter DNA contains both strands of parent DNA, and
- 1 daughter DNA contains both new strands.

2- Semi-conservative model: According to this model, the double-helix of parent DNA is broken. It means that two strands of parent DNA separate. Each of these strands acts as a template for the synthesis of a new strand. So, after replication;

- Both daughter DNAs consist of one strand of parent DNA and one new strand.

3- Dispersive model: According to this model, parent double-helix DNA is broken and dispersed into nucleotides. Then, each nucleotide combines with new nucleotides to generate the four strands of daughter DNAs. So, after replication;

- Both daughter DNAs contain strands which are mixtures of old and new nucleotides.

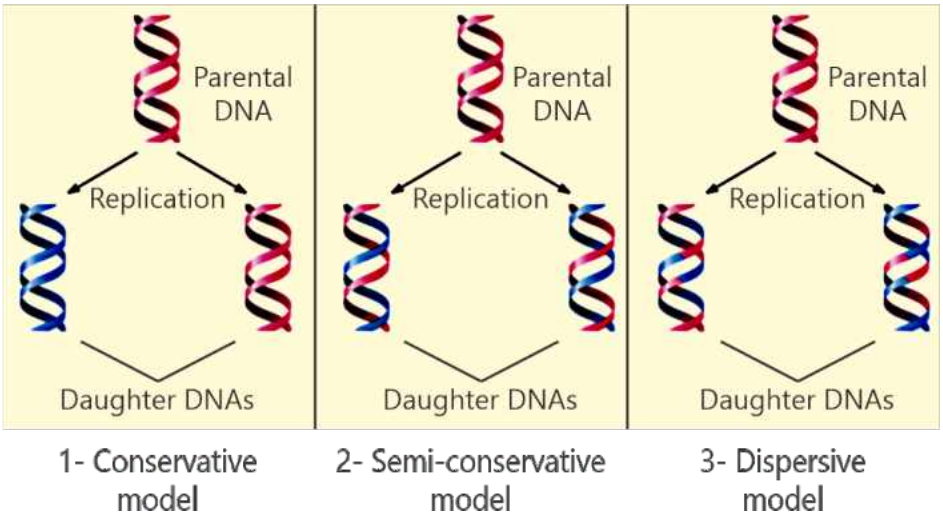


Figure 19.8: Three models of DNA replication

19.3.2- Meselson-Stahl Experiment

In 1958, two American biologists, Mathew **Meselson** and Franklin **Stahl**, evaluated the three models of DNA replication. They proved that the DNA replication is according to semi conservative model.

Experiment

They grew bacteria in a medium containing heavy isotope of nitrogen i.e., ^{15}N. The next generation of the bacteria incorporated ^{15}N in the nitrogen-bases of DNA instead of normal ^{14}N. So, after many generations, the DNA of these bacteria was denser than that of normal bacteria. Then, they transferred the new bacteria (with ^{15}N in DNA) from the ^{15}N medium to a medium that contained ^{14}N. They thought that now these bacteria will make new DNA by using ^{14}N from the medium.

When bacteria divided and increased in number, Meselson and Stahl collected the new DNAs at different intervals. They checked heavier DNA (with ^{15}N) and lighter DNA (with ^{14}N). For this purpose, they separated the heavier and lighter DNAs in the following steps.

1. They dissolved the collected DNA in aqueous solution of cesium chloride and conducted the ultracentrifugation of the solution.
2. The ultracentrifugation created a gradient of cesium density (cesium with more density settled at bottom). DNA strands of different densities were also separated during this ultracentrifugation.
3. Each DNA strand settled at the position where its density was exactly matching the density of the cesium there. The denser DNA with ^{15}N migrated further down as compared to the lighter DNA with ^{14}N.

Observations after Ultracentrifugation

a. First sample was collected at 0 minutes i.e., before any replication. All sample made single sediment of heavy density. It means that all DNA was dense (with ^{15}N).
b. Second sample was collected at 20 minutes i.e., after the bacteria had completed first DNA replication in ^{14}N medium. The sample made a single sediment of intermediate density. It means that all DNA has intermediate density i.e., between ^{14}N-DNA and ^{15}N-DNA.
c. Third sample was collected after 40 minutes when bacteria had completed the second replication. In this sample, DNA made two sediments i.e., one intermediate and one light (equal to that of ^{14}N-DNA).

Tracing radio-isotope labelled DNA in the progeny

An experiment in which a radio-isotope labelled DNA is traced in the progeny of an organism is called a **"pulse-chase experiment."** The experiment has two stages:

- In "pulse" stage, the organism is exposed to the radio-isotope for a brief time. During this step, the organism incorporates radioisotope into its DNA.
- In "chase" stage, the labelled DNA is observed over time by measuring the radioactivity of DNA.

Explanation of Meselson and Stahl Experiment

a. DNA collected before any replication was heaviest because it had both strands made of ^{15}N

b. DNA collected after first replication was intermediate in density because each daughter DNA was a hybrid i.e., one strand was made of ^{14}N and one of ^{15}N.

c. DNA collected after second replication, formed two sediments (one intermediate and one lighter) because each parent DNA contributed one heavy strand to form another hybrid DNA and one light strand to form a light DNA (containing both ^{14}N strands).

On the basis of these results, they claimed that DNA replicates in a semiconservative manner.

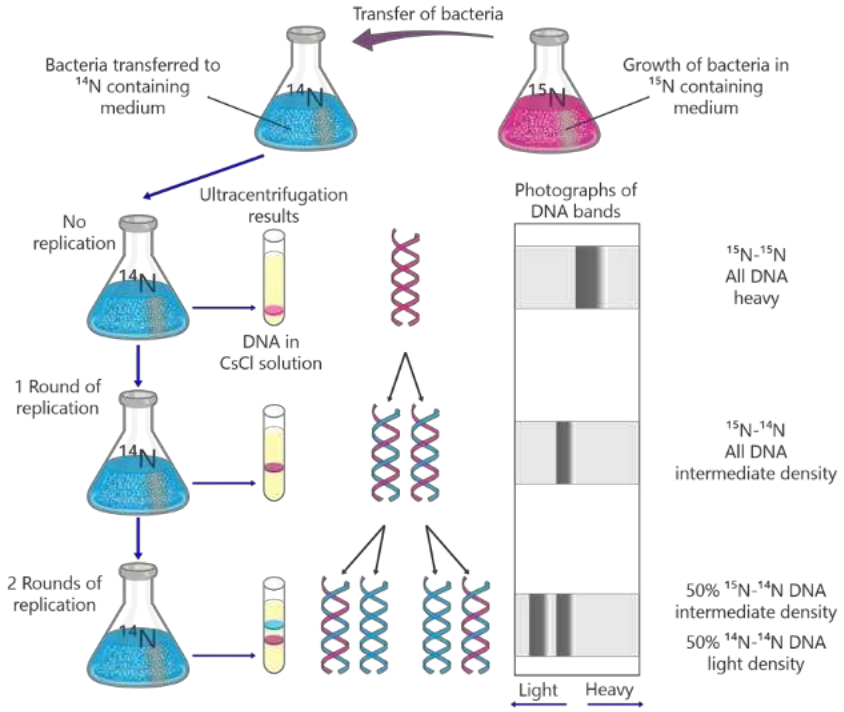


Figure 19.9: Results of Meselson-Stahl experiment

19.3.3- Mechanism of DNA Replication

The basic mechanism of DNA replication is similar in all organisms. The following steps refer to replication in prokaryotes, but eukaryotes also adopt almost the same mechanism.

1- Opening up the DNA double helix

The replication of DNA begins at one or more sites on DNA molecule where there is a specific sequence of nucleotides called a **replication origin site**. In prokaryotes, there is only one replication origin site. But in eukaryotes, there are many replication origin sites along the length of DNA.

i- **Initiator protein** binds to the replication origin site and initiates the process.

ii- At the origin site, the "unwinding" enzymes called **helicases** break the hydrogen bonds between the nucleotide-pairs (base-pairs) of two strands. The stands separate a little and make two Y-shaped structures called **replication forks** (collectively called a replication bubble).

iii- Due to unwinding of the DNA helix, supercoils are produced ahead of the replication fork.

iv- The enzyme **gyrase** (or topoisomerase) attaches ahead of the replication fork. It prevents supercoiling again.

v- **Single-strand binding proteins** bind to single strands and prevent them from re-joining and rewinding.

vi- The two replication forks move away from each other so that replication can occur at both forks. Both single strands act as templates for the next steps.

2- Building a primer

On the exposed strands, new DNA cannot be formed unless some nucleotides are already arranged on template. For this purpose, an enzyme **primase** attaches some RNA nucleotides in front of each template strand at replication origin site. This short fragment of 5-10 RNA nucleotides is called **primer**.

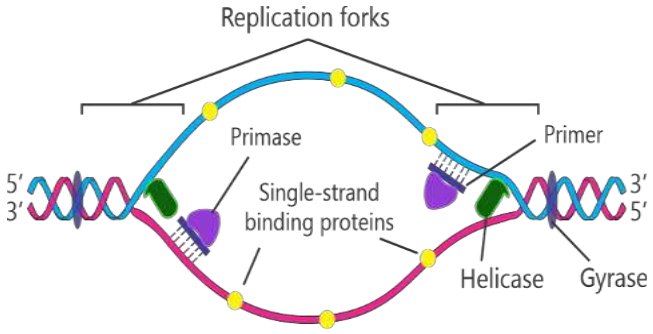


Figure 19.10: Replication – opening up of strands and primer attachment

3- Assembling complementary strands (Extension)

The Main Builder: DNA Polymerase III

The enzyme DNA polymerase adds new nucleotides along both template strands. In prokaryotes, there are DNA polymerase I, polymerase II and polymerase III. But, **DNA polymerase III** is the main enzyme. It grabs free floating nucleotides and matches them to the exposed template strands (A with T, and C with G).

For Information
DNA polymerase I provides a support to polymerase III by replacing the primers at the end of replication. Polymerase II is involved in the repairing process of DNA damages during the life time of a cell.

The Raw Materials

The units used for synthesizing new strands are Deoxyribonucleoside triphosphates (dATP, dCTP, dGTP, dTTP). These molecules have three phosphate groups. As DNA Polymerase III attaches a nucleotide to the strand, it breaks off two phosphates. This action releases energy which is used for the building process.

Tidbit
Why an RNA primer, rather than DNA? Starting chains on exposed templates introduces many errors; RNA marks this initial stretch as "temporary". It makes easy to excise this error-prone later.

One-Way Traffic (5′ to 3′ Direction)

DNA Polymerase III can only add a new nucleotide to the 3′ end of an existing one. First, it adds a nucleotide at 3′ end of primer and then continues adding nucleotides in 5′ to 3′ direction. Because the two original DNA strands run in opposite directions, the enzyme must handle them differently.

Building the Leading Strand

One unit of the enzyme attaches to the template strand that runs toward the opening "fork." Because it is moving in the same direction the DNA is unzipping, it can add nucleotides in one continuous 5′ to 3′ chain. This smoothly growing daughter strand is called the **Leading Strand**.

Tidbit
The leading strand can be extended from one primer alone, whereas the lagging strand needs a new primer for each Okazaki fragment (named for the Japanese scientist who discovered them).

Building the Lagging Strand

The second unit of the enzyme works on the other template strand. However, to maintain the 5′ to 3′ direction, it must move away from the replication fork. As the DNA unzips further, the enzyme has to "jump back" to a new primer to start a new

segment. So, this daughter strand grows discontinuously away from the replication fork. It makes short, separate pieces of DNA called **Okazaki fragments**. Because this strand is built in "stops and starts" rather than one long piece, it is called the **lagging strand**.

4- Termination of replication

The termination of replication occurs at specific sequence of nucleotides called **termination site**. The region of DNA between the origin site and termination site is termed as a replication unit or **replicon**. During termination, DNA polymerase I removes the RNA primer and fills in the gap with DNA nucleotides. It also fills any gaps between Okazaki fragments. The enzyme **DNA ligase** joins the Okazaki fragments of the lagging strand.

For Information
During replication, one strand of the original DNA is conserved, while the daughter strand is synthesized anew. It means that half of the original DNA molecule is conserved in each new generation of DNA.

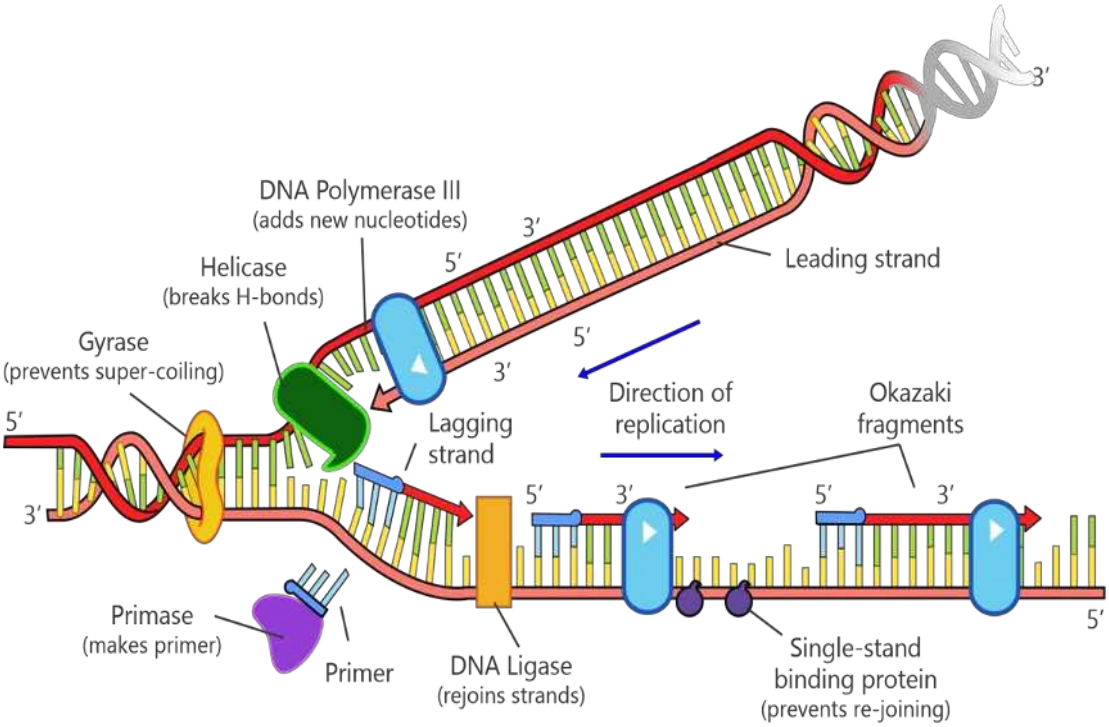


Fig. 19.11 Process of DNA replication

Difference between DNA Replication in Prokaryotes and Eukaryotes

Location: In prokaryotes, replication occurs in the cytoplasm (as they do not have nucleus). In eukaryotes, nucleus is the site for DNA replication.

Polymerases: In prokaryotes, DNA Polymerase I, II, and III work in replication. In eukaryotes, there are at least 14 polymerases (such as alpha, delta, and epsilon).

Initiation and termination sites: In prokaryotes, DNA has a single origin site and a single termination site. In eukaryotes, there are multiple origin and termination sites on each chromosome. DNA replication starts at each origin site simultaneously. Thus, each chromosome has several replicons.

Okazaki fragments: In prokaryotes, the Okazaki fragments are longer (1000 to 2000 nucleotides). In eukaryotes, the Okazaki fragments are short (between 100 and 200 nucleotides).

Speed: Prokaryotes replicate their small DNA very quickly. Eukaryotes have massive amounts of linear DNA. To compensate for this, eukaryotes start replication at thousands of different origin sites along a single chromosome at the same time.

For Information

Biotechnologists use DNA replication in new technologies. For example;

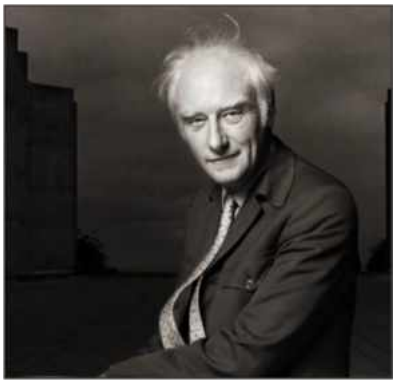
- In Polymerase Chain Reaction (PCR), scientists conduct replication process to make billions of copies of a specific DNA.
- In recombinant DNA technology, scientists cut DNA from different sources. Then, they conduct DNA replication to amplify the DNA molecules.
- In genome sequencing, scientists use DNA replication to make multiple copies of the genome, which are then sequenced to know the genome sequence.
- In diagnostic studies, scientist replicate specific sections of patient's DNA. Then, they search changes or mutations in DNA to know the presence of a genetic disorder.

Stability and Variability of DNA

These are two important properties of a replicating DNA. “Stability” means the ability of DNA to be copied with accuracy. DNA Polymerase III (and eukaryotic polymerases) have a "proofreading" ability to ensure that the sequence of nucleotides is maintained. This consistency is vital because it maintains the specific traits of an organism. It also ensures that cells function correctly.

While stability is the rule, “variability” is an exception. Occasionally, a permanent change in the DNA sequence occurs, known as a **mutation**. These mutations can be caused by errors during replication. Environmental factors can also cause mutations. Variability is the primary source of new traits. It allows populations to adapt to changing environments and is the raw material for evolution.

19.4- Gene Expression


Francis Crick

You know that a gene is a specific sequence of nucleotides along the DNA strand which directs the synthesis of a polypeptide (protein). In other words, a gene expresses itself by making a protein or an enzyme that controls the development of a specific trait.

All organisms use the same basic mechanism of expressing genes. English biologist, Francis Crick used the term **"Central Dogma"** for this mechanism. The central dogma states that information passes from the genes (DNA) to an RNA copy of the gene, and the RNA copy directs the sequential assembly of a chain of amino acids. In simple words, the central dogma is;

DNA → RNA → Protein

It means that the flow of genetic information is one-way. Once the information has gone into a protein, it never goes back.

The central dogma consists of two steps i.e., transcription (synthesis of RNA from DNA) and translation (synthesis of a protein by using the information present in RNA).

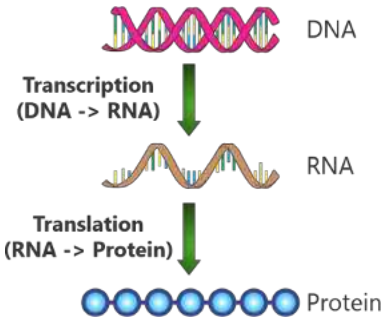


Figure 19.12: Central dogma

19.4.1- Genetic Code

After the discovery of the structure of DNA (in 1953) and the idea of central dogma (in 1957), the essential questions were;

- How can DNA, a substance containing only four different nucleotides, store genetic information for making thousands of specific proteins?
- If DNA has genetic information in the form of a genetic code, what is the nature of this code?
- If genetic code is in the form of the order of nucleotides in a DNA, how does it specify the order of amino acids in a polypeptide?

These questions started a race among biologists. The following were the main achievements, in this regard.

Work of Francis Crick

In 1961, Francis **Crick** and his colleagues presented an idea; "the genetic code consists of a series of blocks of information called **codons**. Each codon specifies one amino acid of the polypeptide to be synthesised". They further thought that;

- If a sequence of single nucleotides specifies a single amino acid, only four different amino acids could be encoded.
- Even a sequence of 2 nucleotides would make 16 (4 x 4) codons and they would specify 16 amino acids.

For Information
The information in DNA for the synthesis of proteins is called as genetic code. The genetic code consists of a series of codons consisting of 3-nucletotide.

They reached to a conclusion that one codon should be a sequence of 3 nucleotides. This 3-nucleotide sequence specifies a particular amino acid. The 4 nucleotides can be arranged in 64 (4 x 4 x 4) different combinations which are more than enough to code for the 20 amino acids.

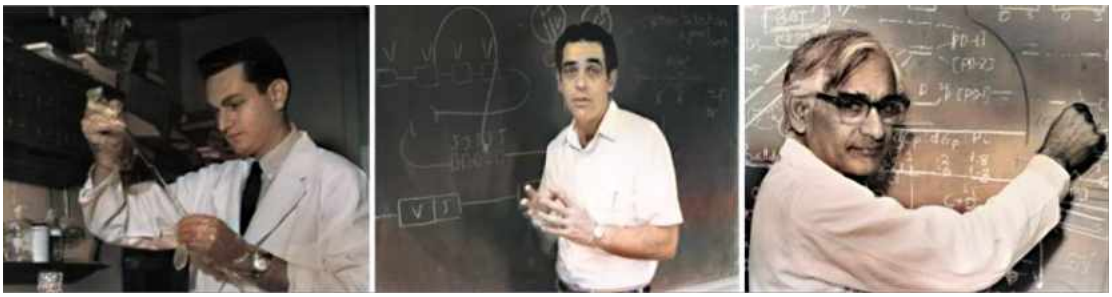
According to the conclusions of Crick, the genetic code consists of continuous sequences of three nucleotides. In other words, it is a **triplet code**. When a gene works, the codons present on the DNA are transcribed in the form of complementary codons of mRNA. So, the TAC codon on DNA would mean AUG codon on mRNA. In general, the term "codon" refers to the codon of mRNA. However, in some cases, the term "codon" may also be used to refer to codons of DNA, especially when discussing mutations in DNA.

Work of Marshall Nirenberg, Philip Leder and Har Gobind Khorana

In 1961, the American biochemist, Marshall **Nirenberg** made a poly-U mRNA. This mRNA consisted of uracil nucleotides only. He added this poly-U mRNA to the cytoplasm of bacteria. He observed that the bacteria produced a polypeptide consisting of many phenylalanine amino acids. He concluded that the codon UUU specifies phenylalanine. Nirenberg and another American geneticist Philip **Leder** tested all triplet codons. They determined the amino acids specified by 47 codons. Har Gobind **Khorana** (a scientist who completed his studies in Government College, Lahore) determined the amino acids specified by the remaining 17 triplet codons.

Out of the total 64 codons, three codons (UGA, UAG, and UAA) are **stop codons** or **nonsense codons**. If any of these is present at the end of mRNA, it does not specify any amino acid and stops the protein synthesis. The codon AUG acts as **start codon**. It is present at the beginning of all genetic codes. It specifies the amino acid methionine.

The 1968 Nobel Prize in Physiology & medicine awarded to Nirenberg (work on genetic code), Har Gobind Khorana (synthesis of nucleic acids), and Robert Holley (discovering the chemical structure of transfer-RNA).



(Left to Right)- Marshal Nirenberg, Philip Leder, Har Gobind Khorana

19.4.2- Characteristics of Genetic Code

1- Genetic code is universal: The genetic code is same in almost all organisms. For example, AGA specifies arginine in bacteria, in humans and all other organisms. Due to this characteristic, genes can be transferred among organisms.

However, there are a few exceptions to this universality. For example, genetic code in the DNA of mitochondria is slightly different from the genetic code in the DNA of chromosomes. It reflects the independent evolutionary history of mitochondria. Similarly, some organisms (e.g., ciliates) and viruses (e.g., bacteriophages) use different genetic codes for the same amino acids.

For Information
The exceptions to the universality of the genetic code demonstrate that while the standard genetic code is widely used, there is still room for variation and adaptation in the genetic code.

2- Genetic code is triplet: The genetic code in DNA is a sequences of three nucleotides (codons). Due to this characteristic, the amount of information present in DNA balances the information required to specify 20 kinds of amino acids.

Analyse & Interpret
Interpret how many types of t-RNA molecules are necessary for a living cell, if the genetic code is a triplet code.

3- Degeneracy of genetic code: Degeneracy of genetic code means that multiple codons can specify the same amino acid. For example, four different codons (AUG, GUG, UUG, and CUG) code for the same amino acid i.e., methionine. Characteristic is important for mutation tolerance and evolutionary flexibility.

4- A codon does not have punctuations: A codon is read continuously. It means that it is without punctuation (gaps) between the three-nucleotides of a codon.

5- Genetic code is non-overlapping: It means that each triplet codon is read independently of the adjacent codons. For example, the series of AUGAGCGCA codons cannot be read as AUG/UGA/GAG etc. These will only be read as AUG/AGC/GCA.

Table: Codons (on mRNA) and their specific amino acids (or stop functions)

SECOND NUCLEOTIDE

FIRST NUCLEOTIDE	U		C		A		G		THIRD NUCLEOTIDE
U	UUU	Phenylalanine	UCU	Serine	UAU	Tyrosine	UGU	Cysteine	U
	UUC		UCC		UAC		UGC		C
	UUA	Leucine	UCA		UAA	Stop	UGA	Stop	A
	UUG		UCG		UAG	Stop	UGG	Tryptophan	G
C	CUU	Leucine	CCU	Proline	CAU	Histidine	CGU	Arginine	U
	CUC		CCC		CAC		CGC		C
	CUA		CCA		CAA	Glutamine	CGA		A
	CUG		CCG		CAG		CGG		G
A	AUU	Isoleucine	ACU	Threonine	AAU	Asparagine	AGU	Serine	U
	AUC		ACC		AAC		AGC		C
	AUA		ACA		AAA	Lysine	AGA	Arginine	A
	AUG	Methionine (Start)	ACG		AAG		AGG		G
G	GUU	Valine	GCU	Alanine	GAU	Aspartate	GGU	Glycine	U
	GUC		GCC		GAC		GGC		C
	GUA		GCA		GAA	Glutamate	GGA		A
	GUG		GCG		GAG		GGG		G

19.5- Transcription

It is the first step of gene expression. During transcription, a complementary RNA is synthesised from DNA. Only one strand of DNA, called the **template strand**, is transcribed. The mRNA synthesised in transcription is complementary to the template strand of DNA. The other strand of DNA that is not transcribed is called the **coding strand** because it has the same sequence as the transcribed mRNA, except T takes the place of U. The following are the main phases of the transcription of mRNA.

For Information
The template strand of DNA is also known as the antisense (–) strand and coding strand as the sense (+) strand.

1- Initiation Phase

In this process, no primer is needed. An enzyme RNA polymerase binds at template DNA at a special nucleotides sequence. This sequence is called **promoter**. It is not itself transcribed by the enzyme. For example;

- In prokaryotes, a promoter **TTGACA** is present 35 nucleotides before the start point of gene. So, it also called –35 sequence.
- In prokaryotes, another promoter **TATAAT** is 10 nucleotides before the start point of gene. It is also called –10 sequence.

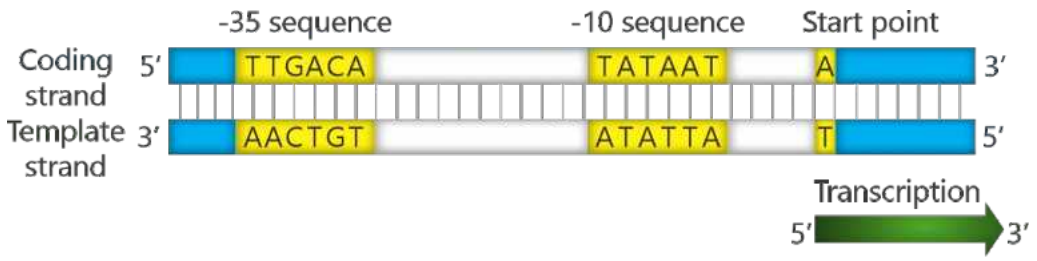


Figure 19.13: Promoters in prokaryotes

- In eukaryotes, a promotor **TATAAA** (called **TATA box)** is 25 nucleotides before the start point. It is also called –25 sequence.
- In eukaryotes, a promoter **GGCCAATCT** (called **CAAT box)** is 70 nucleotides before the start point. It is also called –70 sequence.

For Information
Prokaryotes have only one RNA polymerase, which synthesises all 3 kinds of RNA. Eukaryotes have three different RNA polymerases: RNA polymerase I synthesizes rRNA in the nucleolus; RNA polymerase II synthesizes mRNA; and RNA polymerase III synthesizes tRNA.

RNA polymerase consists of five subunits:

- two **α subunits** bind with regulatory proteins,
- a **β′ subunit** binds with template DNA,
- a **β subunit** binds with RNA nucleotides, and
- a **σ subunit** recognizes the promoter and initiates synthesis.

When RNA polymerase attaches to promoter, its **σ subunit** is removed. The other four subunits (core enzyme) catalyse the process. They unwind the DNA helix. They breakdown nucleotide-pairs (base-pairs). It results in the formation of a bubble-like structure, the **transcription bubble**. The assemblage of RNA chain occurs in this bubble.

2- Elongation Phase

Inside the transcription bubble, RNA polymerase begins making a chain of complementary ribonucleotides in front of template DNA strand. It is done in 5′ to 3′ direction. It usually starts with A or G ribonucleotide. Here, the nucleotides available for attachment are in the form of ribonucleoside triphosphates (rNTP). When an rNTP is added, its two phosphates detach with the release of energy, and ribonucleotide is attached to the chain.

The first 12 bases of new mRNA strand make a temporary helix with template DNA strand. Due to it, the position of the 3′ end of the mRNA remains stable. So, it can receive an incoming ribonucleotide. The RNA-DNA helix rotates each time a nucleotide is added. So, the 3′ end of RNA stays at the catalytic site of RNA polymerase. The

transcription bubble moves down the DNA. The growing mRNA strand protrudes from the bubble. After the transcription bubble passes, the transcribed DNA is re-wound.

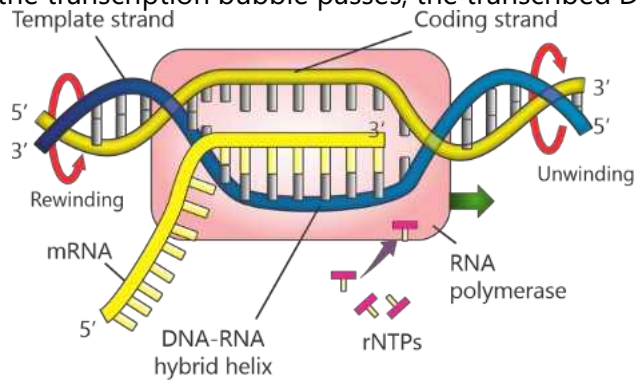


Figure 19.14: Elongation phase of transcription

3- Termination Phase

At the end of a gene on template DNA. There is a stop codon. After this stop codon, there is a series of **stop region** in the form of GC nucleotide-pairs (base-pairs) followed by a series of AT pairs. When these nucleotide-pairs are transcribed into mRNA, hydrogen bonds are formed within the strand of mRNA. This intra-strand nucleotide-pairing (base-pairing) creates a loop called **GC hairpin** with a tail of four or more U ribonucleotides. The GC hairpin causes the RNA polymerase to pause. The pairing of four U of mRNA of with A of DNA is the weakest. It cannot hold the RNA-DNA hybrid strands together during the long pause. So, the RNA strand dissociates from DNA stand within the transcription bubble, and transcription stops.

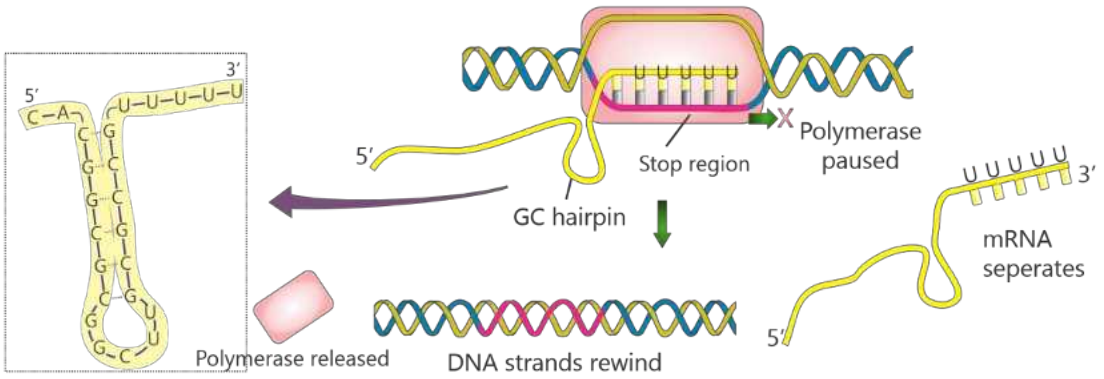


Figure 19.15: Termination phase of transcription

Post-Transcriptional Modifications

As the prokaryotes have no nucleus and transcription happens in cytoplasm, their new mRNA does not need to travel anywhere to start translation. On the other

hand, in eukaryotic cells the new mRNA has to travel from nucleus to cytoplasm. For this journey, the new mRNA undergoes the following modifications.

1- Addition of 5′ Cap: The first nucleotide (at the 5′ end of mRNA) is A or G. At the terminal 5′ end of A or G a methylated GTP is attached. This bond is formed between 5′end of terminal A or G and 5′end of GTP. This structure is called a 5′ cap. It protects the mRNA from nucleases and phosphatases during its journey.

2- Addition of 3′ Poly-A tail: At the 3′end of mRNA, a small chain of about 250 A nucleotides is added. It is called 3′ poly-A tail. It protects the mRNA transcript from degradation by nucleases.

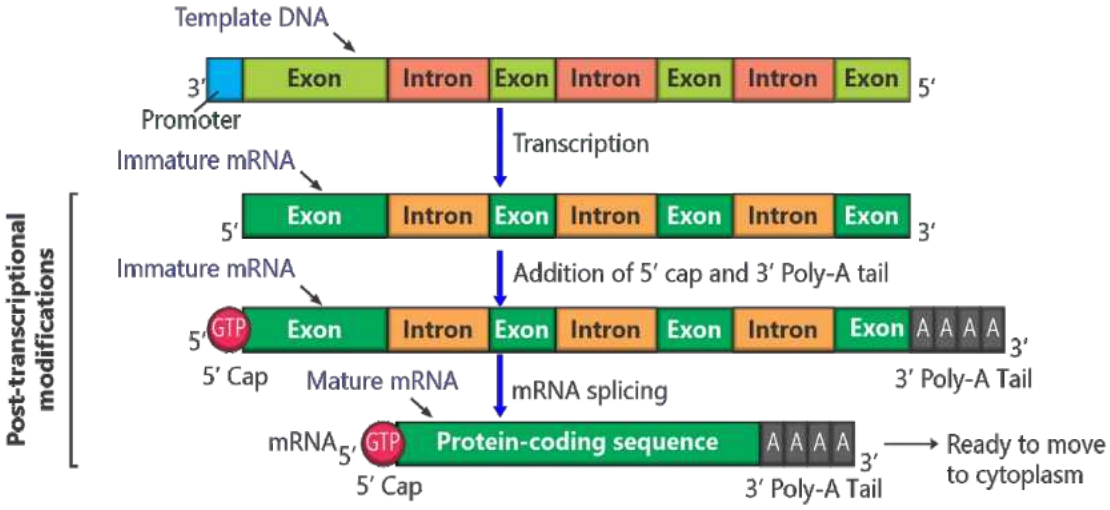


Figure 19.16: Post-transcriptional changes in eukaryotes

3- Splicing: The new mRNA is immature because it contains introns (regions of DNA or mRNA that do not code for proteins) and exons (regions of DNA or mRNA which are "expressed" or translated into a protein). mRNA splicing is the process by which introns are removed from the immature mRNA. After splicing, the remaining exons are joined. So, the mature RNA consists of coding segments (gene) only.

19.6- Translation

In translation or protein synthesis, the information present in an mRNA transcript is used to direct the sequence of amino acids to produce a polypeptide.

Activation of Amino Acids

Before protein synthesis, the assemblage units are prepared. Specific tRNAs attach with specific amino acids and make **aminoacyl-tRNAs (aa-tRNA)**. The enzyme **aminoacyl-tRNA synthetase** carries out this reaction. There are 20 kinds of this enzyme. Each kind binds to a specific amino acid. It also recognizes and binds to the tRNA with anticodons for that amino acid. The aa-tRNA delivers the amino acid to the ribosome for incorporation into the polypeptide chain.

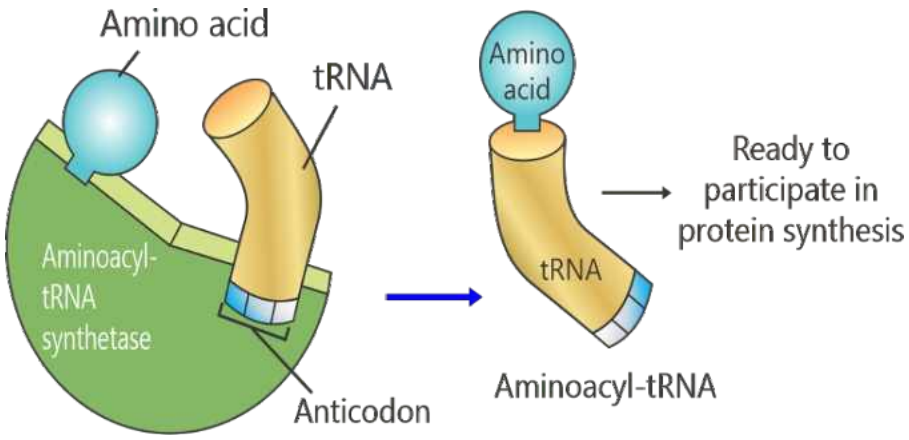


Figure 19.17: Activation of amino acids

1- Initiation Phase

In this phase, an **initiation complex** is formed in the following steps:

1. The aa-tRNA carrying the first amino acid binds to the smaller ribosomal subunit. An enzyme called **initiation factor** controls this binding. In prokaryotes, the first amino acid is modified methionine i.e., N-formyl-methionine. While in eukaryotes, it is non-modified methionine
2. Another initiation factor attaches the 5′ end of mRNA with smaller ribosomal subunit. It positions the anticodon of aa-tRNA over the first codon (AUG) of mRNA.
3. The larger ribosomal subunit is also placed upon smaller subunit.

The **initiation complex** formed in this way consists of both ribosomal subunits, first aa-tRNA, mRNA, and the enzyme.

The site of ribosome where first aa-tRNA is placed consists of;

- A **Peptidyl site** (P site) – where peptide bonds will form between amino acids.
- An **aminoacyl site** (A site) – where next aa-tRNA will bind.

- An **exit site** (E site) – where empty tRNAs will exit the ribosome.

With respect to mRNA, the three sites are oriented in 5′ to 3′ as E-P-A, because ribosomes move toward the 3' end of mRNA.

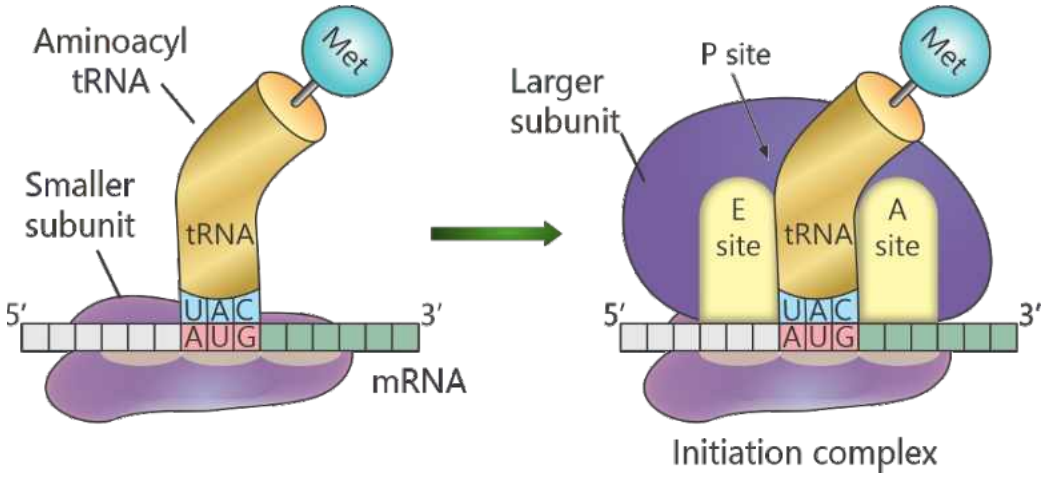


Figure 19.18: Initiation phase of translation

2- Elongation Phase

In the initiation complex, the P-site of ribosome is occupied by first aa-tRNA (with its anticodon matching the mRNA codon). The next codon of mRNA is exposed at A site. Elongation happens in the following steps;

1. The next aa-tRNA with matching anticodon and matching amino acid binds to A site with the help of an enzyme, the **elongation factor**.
2. Another enzyme **peptidyl transferase** removes methionine from first aa-tRNA present on P site and attaches it by a peptide bond to the amino acid of the second aa-tRNA present at A site.
3. The ribosome moves three nucleotides along the mRNA in 5´ - 3´ direction. This movement is called **translocation**.
4. As a result of the previous step, the aa-tRNA carrying the chain of two amino acids is shifted from A site to P site. The new codon of mRNA is also exposed at A site. Moreover, the empty tRNA comes to E site to leave the ribosome.
5. These steps are repeated again and again until the stop codon is reached at A site.

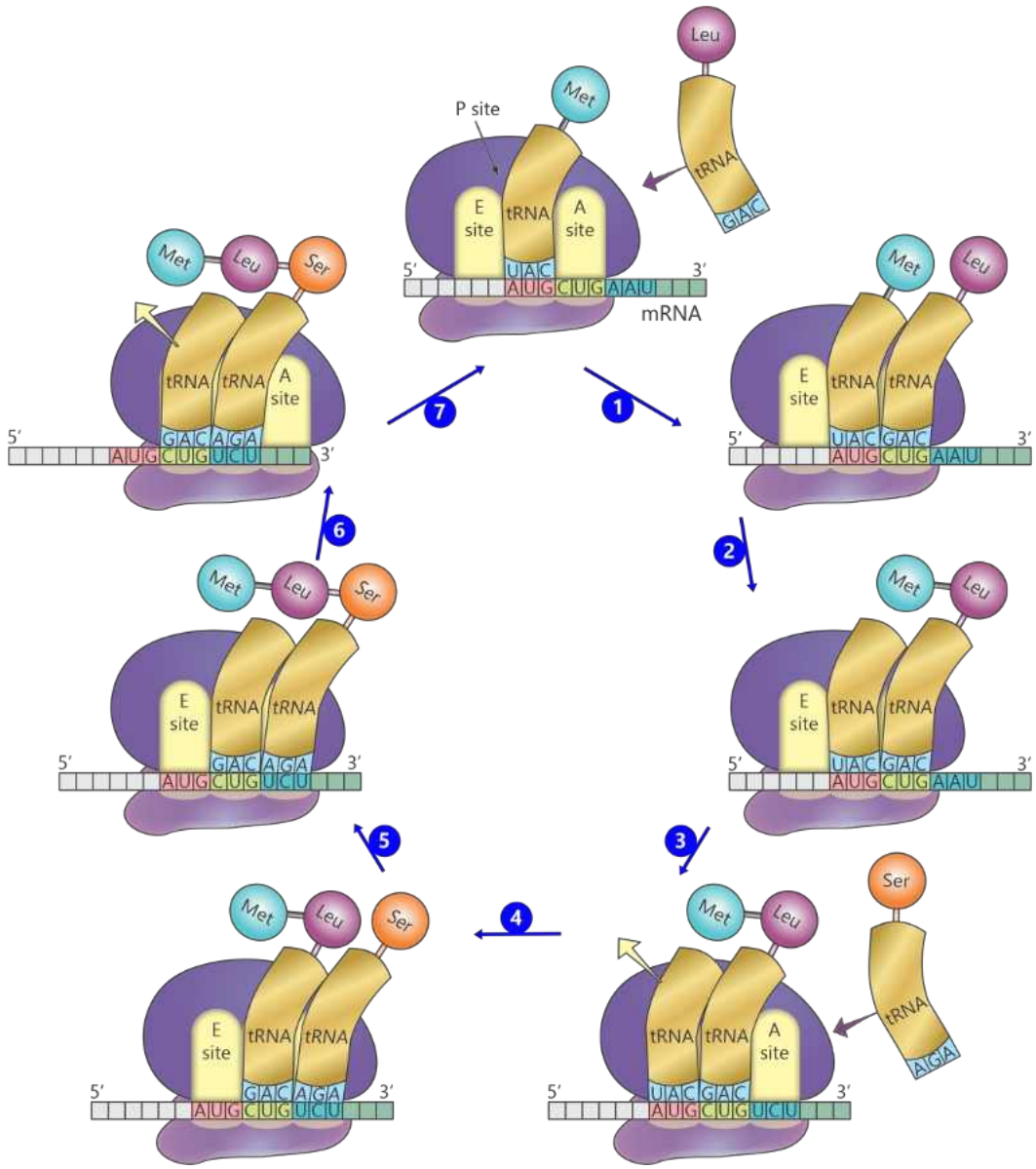


Figure 19.19: Elongation phase of translation (1- second aminoacyl-tRNA binds to A site; 2- first amino acid is removed from first tRNA and attached to the amino acid of the second aminoacyl-tRNA; 3- ribosome moves three nucleotides in 5´ - 3´ direction; 4- third aminoacyl-tRNA binds to A site; 5- chain of first two amino acids is removed from the tRNA at P-site and attached to the amino acid of the third aminoacyl-tRNA; 6- ribosome again moves three nucleotides and next codon of mRNA is exposed at A site; 7- the process is repeated for all codons)

3- Termination Phase

When a stop codon is exposed at A site, no aminoacyl-tRNA comes to A-site (because there is no anticodon of the stop codons). Stop codon at A site is recognized by a protein called **release factor**. It releases the polypeptide from the tRNA present at P site. In this way, the process of translation is terminated. The tRNA is also released from ribosome and ribosomal subunits separate from the mRNA.

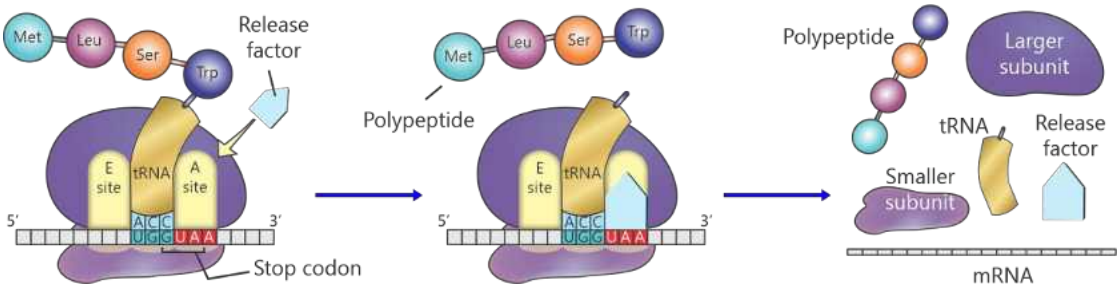


Figure 19.20: Termination phase of translation

19.6.1- Difference between Translation in Prokaryotes and Eukaryotes

	Prokaryotes	Eukaryotes
1	Prokaryotic genes lack introns.	Most eukaryotic genes possess introns.
2	After transcription, the mRNA immediately undergoes translation. There is no post-transcriptional modification in mRNA.	The mRNA is modified before its translation: introns are cut out; a 5′ cap is added; and a 3′ poly-A tail is added.
3	Translation occurs in cytoplasm.	Translation occurs in cytoplasm as well as in rough endoplasmic reticulum.
4	Ribosomes are smaller in size (70S).	Ribosomes are larger in size (80S).
5	The initial amino acid is modified i.e., N-formyl methionine.	The initial amino acid is methionine. It is not modified.

19.7- Regulating Gene Expression

All of the genes in an organism are not expressed simultaneously. Instead, different genes are expressed at different times. Similarly, all types of cells contain the same genome but all genes are not turned ON in all cells. Instead, specific genes are turned ON in each cell type. Turning the genes ON and OFF and the timing of their expression are controlled by **regulator proteins** and the process is called regulation of gene expression.

Importance of Regulation of Gene Expression

Regulation of gene expression enables the cells to respond to environmental conditions and to prepare protein when needed. In multicellular organisms, gene regulation is the basis for cellular differentiation and morphogenesis. It leads to the creation of different cell types. In different cell types, the genome is the same but different genes are expressed according to the type.

Methods of Regulation of Gene Expression

Regulation of gene expressions occurs at several levels. For example, at transcription level, the rate of transcription is regulated. At post-transcriptional level, the modification of mRNA is controlled. At translation level, the rate of translation is regulated. At post-translational, the modification of protein (its folding, stability etc.) is regulated. The important methods of regulation of gene expression include;

1- Regulation at Transcriptional Level

The genes have special regulatory sequences. Specific regulatory proteins bind to these sequence. These proteins can take two types of actions;

a. **Negative gene regulation:** Some regulatory proteins, called **repressors** bind to some pieces of DNA, called **operators**, present near the promoter. Here, the repressors block RNA polymerase and stop or reduce transcription.

b. **Positive gene regulation:** Some regulatory proteins, called **activators (or inducers)** bind to the operators. Here, the activators start transcription or increase its speed by helping in the binding of RNA polymerase to the promoter.

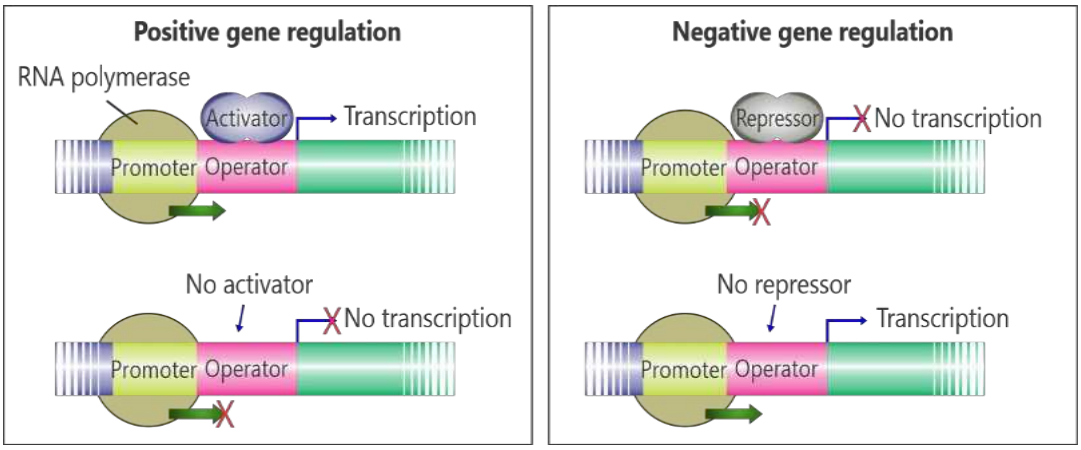


Figure 19.21: Positive and negative gene regulation by regulatory protein

2- Regulation during Splicing

In eukaryotes, mRNA is modified before its export to cytoplasm. Genes can also be regulated (tuned ON or OFF) during mRNA splicing (removal of introns and joining the exons). Specific regulatory proteins in specific cell types control splicing. These proteins remove the pieces of mRNA (introns or exons) in different ways. So, mRNAs of different lengths are produced. It results in the production of more than one polypeptide from a single gene.

Tidbit
Liver cells express specific genes which make the enzyme alcohol dehydrogenase. This enzyme breaks alcohol down into a non-toxic molecules. The neurons of brain also have these genes but they keep these genes unexpressed or "turned OFF".

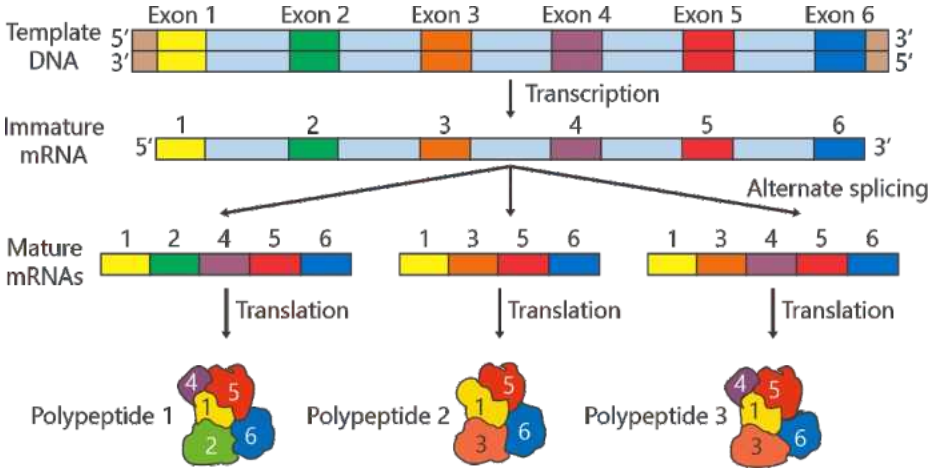


Figure 19.22: Alternate splicing and different gene products

19.8- Mutations

A change in the content of genetic message is called mutation. Mutations may range from small (e.g., a change in a single or few nucleotides) to large (e.g., a change in a large segment or whole chromosome). There may be natural mutations and induced mutations.

Natural mutations occur spontaneously as a result of sequencing errors or DNA damage during replication. These mutations are a part of the process of evolution and can result in beneficial, neutral, or harmful effects on organism.

Mutations – Contributors in Evolution
The ultimate sources of all genetic variation are mutation are recombination. These processes create new DNA sequence (allele) and combination of alleles. The new alleles form new traits which may be advantageous, harmful or neutral. Natural selection acts upon the variations created by new traits and the allelic frequencies of advantageous traits increase. It leads of evolution in the long run.

Induced mutations are caused by external factors such as exposure to mutagen substances or radiation. Such mutations can also result from exposure to chemicals in the environment, including certain pesticides and industrial pollutants, or from medical treatments such as chemotherapy. Induced mutations can have serious health effects on an organism.

For Information
The agents that cause mutations are called mutagens while the organism or cell in which mutation occurs is called mutant.

On the basis of the size of mutations, we can classify them in two main types i.e., point mutations and chromosomal mutations.

19.8.1- Point Mutations

The mutations involving the change in only one or a few nucleotides, are called point mutations. The following are the common examples:

1. **Substitutions:** These mutations involve exchange of a single nucleotide for another e.g., during replication, A may be substituted with G or C is substituted with T.
2. **Insertions**: It is the addition of one or more extra nucleotides into the DNA. These mutations usually occur due to errors during replication.
3. **Deletions:** Such mutations remove one or more nucleotides from the DNA.

For Information
Gene mutation is a general term for any change within the chemical structure of a single gene. It is a change at the "molecular level" (the nucleotide sequence) rather than at the "chromosomal level" (large chunks of DNA). **Point mutation** are the most common gene mutation.

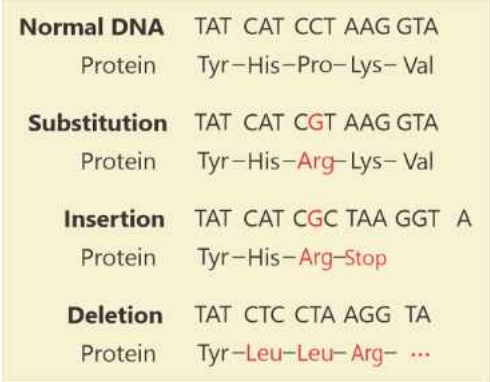


Figure 19.23: Point mutations

Sources of Point Mutations

1- Physical Damage to DNA

Ionizing Radiation: High-energy forms of radiation, such as X rays and gamma rays, are highly mutagenic. Such radiation change atoms into free radicals which may break phosphodiester bonds of DNA strand.

Ultraviolet Radiation: DNA strongly absorbs UV radiation. When two adjacent pyrimidine nucleotides (C and T) absorb UV radiation, a double covalent bond forms between them. It can result in complete replication blockage.

2- Mutagen Chemicals

Many point mutations are caused by mutagen chemicals. Some of these chemicals resemble nucleotides and are incorporated into DNA. Such DNA cannot be

properly transcribed. Some chemicals remove the amino group from adenine or cytosine, causing them to mis-pair; and some chemicals add hydrocarbon groups to nucleotides, also causing them to mis-pair. Such mutagen chemicals are commonly used in laboratories, and sometimes released into the environment (e.g., mustard gas).

3- Spontaneous Point Mutations

Many point mutations occur spontaneously. Sometimes nucleotides spontaneously shift to alternative positions. During replication, DNA polymerase pairs a different nucleotide with the shifted nucleotide. Sometimes, the nucleotide sequences misalign when homologous chromosomes pair. It causes a portion of one strand to loop out. The cell cuts out the loop and it results in a deletion of several hundred nucleotides from one of the chromosomes.

Minor Mutations in Humans

Single nucleotide polymorphisms (SNPs): These mutations involve a single nucleotide. These are very common and can be harmless or have a subtle effects. For instance, a single base change in a gene can make a person unable to digest milk (lactose).

Insertions and deletions: These mutations add or remove one or more nucleotides. For example, the common cause of **Cystic Fibrosis** is a deletion of just three nucleotides in a gene. This deletion causes a specific amino acid to be left out, resulting in a protein that doesn't fold correctly, leading to lung and digestive issues.

Silent mutations: These mutations do not change the amino acid sequence of protein, for example, a DNA code changes from GGC to GGU. Because of the "degeneracy" of the genetic code, both of these codes translate to the same amino acid: Glycine.

Missense mutations: These mutations change only one amino acid, which can affect protein function. For example, a single nucleotide change (A to T) in the gene for haemoglobin causes one amino acid (Glutamic acid) to be replaced by another (Valine). This single "wrong" amino acid causes red blood cells to lose their round shape and causes sickle cell anaemia

Nonsense mutations: These mutations create a premature stop codon in the protein sequence. It results in shortened and non-functional proteins. For example, in Duchenne Muscular Dystrophy, a nonsense mutation creates a premature stop signal in the middle of the gene. The resulting protein is too short and cannot function, leading to progressive muscle weakness.

Why are most mutations harmful?

Most mutations disrupt the normal functioning of genes and prove harmful. For example;

- A mutation that changes a single amino acid in the haemoglobin protein can lead to sickle cell anaemia, a debilitating and potentially life-threatening disease.
- Mutations can also disrupt gene regulation and can alter developmental processes and contribute to the development of various diseases, such as cancer.
- Some mutations can create non-functional or unstable proteins that can be harmful to the organism e.g., phenylketonuria.

Genetic Disorders Caused by Point Mutations

The disorders that result from a change in the DNA sequence can be inherited in an autosomal dominant, autosomal recessive, or X-linked manner. The following are two important genetic disorders caused by single-gene mutations.

1- Sickle Cell Anaemia

Sickle cell anaemia is due to a recessive gene of abnormal β chain of haemoglobin. This gene is present on chromosome 11. The recessive gene has a point mutation in which the DNA nucleotide T is substituted by A. So, the codon CTC of glutamic acid is converted to CAC, which is for valine. If a child receives both recessive (mutant) alleles, he produces abnormal β chains (with glutamic acid replaced by valine). The abnormal haemoglobin carries lesser than normal oxygen. Due to this abnormal haemoglobin, RBCs also become sickle shaped. Major symptoms of sickle cell anaemia include pain, fatigue, shortness of breath, jaundice, and an increased risk of infections.

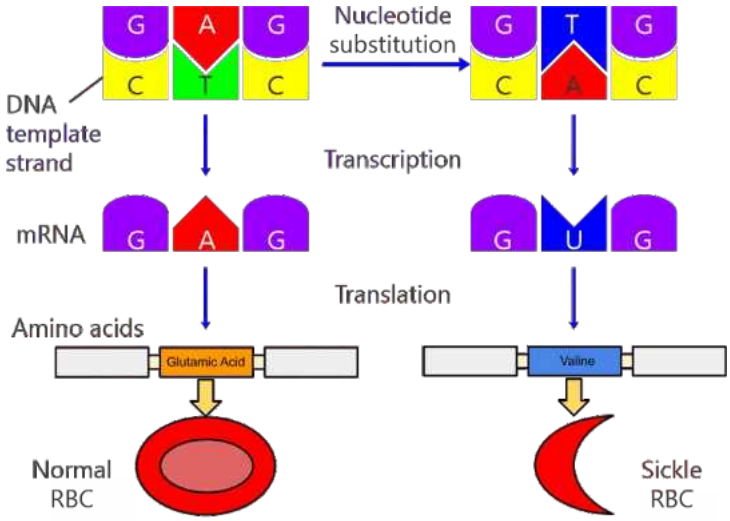


Figure 19.24: Point mutation, causing sickle cell anaemia

The treatments for sickle cell anaemia include pain management with nonsteroidal anti-inflammatory drugs (NSAIDs) and opioids etc. Blood transfusions can help to increase the number of healthy RBCs. Bone marrow transplants can provide a long-term cure. Regular monitoring and preventative measures such as vaccinations and antibiotics can help to prevent complications.

2- Phenylketonuria

It is a genetic metabolic disorder. A normal gene present on chromosome 12 produces an enzyme of liver called **phenylalanine hydroxylase** (PAH). This enzyme coverts amino acid phenylalanine into tyrosine. Point mutation (e.g., substitution or

deletion of a nucleotide) makes the allele recessive. If a child inherits both recessive alleles, PAH enzyme is not produced properly and its activity is reduced. As a result, phenylalanine can build up to toxic levels in the blood and other tissues. This condition is called phenylketonuria (PKU).

Major symptoms include intellectual disability, seizures, behavioural problems, mental disorder, microcephaly (smaller head size), and musty odour in breath, skin, and urine. Excessive phenylalanine can also cause brain damage.

Individuals with PKU have to avoid foods that are high in phenylalanine (e.g., meat, dairy products, eggs, and some grains). The blood phenylalanine levels are monitored regularly to adjust the diet. Some medications contain synthetic PAH enzyme. These are used to lower blood phenylalanine levels. Patients are also given nutritional supplements to ensure they are getting enough vitamins and minerals that may be lacking in the low-phenylalanine diet.

Possible ways to treat genetic diseases

Gene therapy: It involves correcting or replacing the mutated gene with a functional gene.

Cell therapy: For example, in type 1 diabetes, the pancreatic beta cells that produce insulin do not function. Cell therapy involves replacing these cells with healthy ones through transplantation.

Pharmacogenomics: This involves tailoring drug treatments to an individual's genetic makeup. By analysing a patient's DNA, doctors can identify the best drug for them.

Genetic counselling: For individuals with a family history of genetic diseases, genetic counselling can provide valuable information and support. This includes genetic testing, education on the disease, and guidance on family planning and reproductive options.

19.8.2- Chromosomal Mutations

These are large-scale mutations and involve changes in the position of genes, or changes in the structure or number of chromosomes. Such mutations are usually spontaneous. The following are the types of chromosomal mutations.

1- Changes in Chromosome Structure

Chromosomal Rearrangements: Chromosomes can undergo physical alterations. Such changes may involve deletions, duplications, inversions, and translocations. Usually, these changes are caused by a breakage in the DNA double helices at two different locations. In translocations, a segment of one chromosome becomes part of another chromosome. In inversions, the orientation of a portion of a chromosome is reversed.

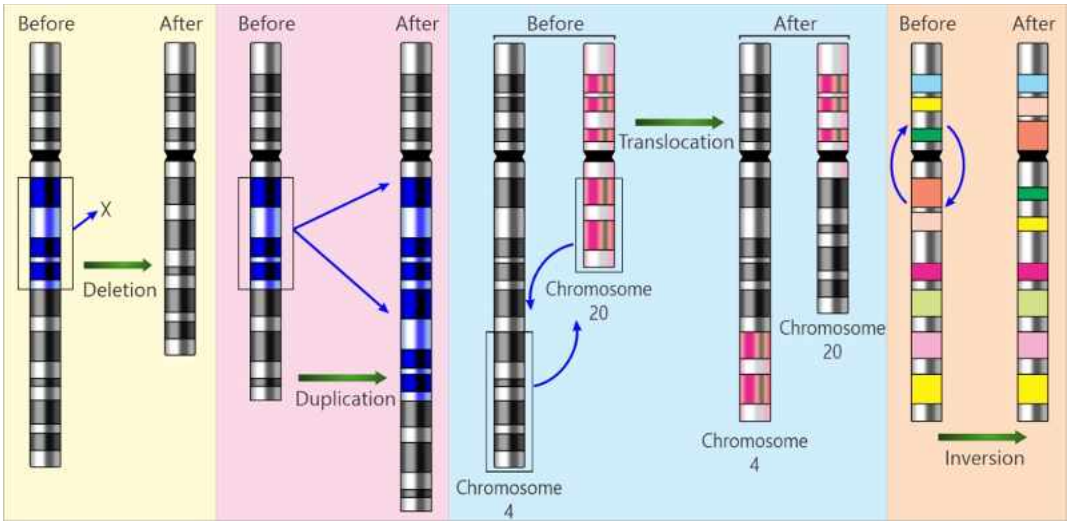


Figure 19.24: Chromosomal rearrangements

Insertional Inactivation: Many small segments of chromosomes can move from one location to another in the genome; These mobile bits of DNA are called **transposons** or **jumping genes**. Transposons select their new locations at random. Some transposons are able to be inserted into genes and inactivate the genes. This form of mutation is common in nature. Many human gene disorders are the result of transposition. For example, a human transposon gets inserted into clotting factor IX gene on X chromosome. It places a stop codon in the middle of the gene. It results in X-linked haemophilia.

2- Changes in Chromosome Number (Chromosomal Aberrations)

Occasionally, sister chromatids fail to separate properly in meiosis, leading to the gain or loss of a chromosome in a gamete. This condition, called **non-disjunction**, can result in individuals with severe abnormalities if the affected gamete forms a zygote. Nondisjunction can happen in autosomes or in sex chromosomes. The following are the major abnormalities that arise due to non-disjunction.

Tidbit

There is a much greater chance of non-disjunction to accumulate over time in eggs than in sperm.

The reason is that all of the future eggs have developed to the point of prophase I by the time a female is born. It means that when she has children, her eggs are as old as she is. In contrast, men produce new sperm daily. Therefore, the age of mother is more critical than that of father.

1- Down Syndrome

In this developmental defect, the total chromosome number is 47 (2n+1). It was first described in 1866 by J. Langdon **Down**; that's why it is called Down syndrome (formerly "Down's syndrome"). About 1 in every 750 children have Down syndrome. It is caused by non-disjunction in the autosome pair 21. This non-disjunction can happen during the formation of egg in women. The 21st chromosomal pair fails to segregate properly during meiosis and results in the formation of an egg having 24 chromosomes. The fertilization of such egg by a normal sperm produces a child (male or female) with **trisomy** having 47 chromosomes.

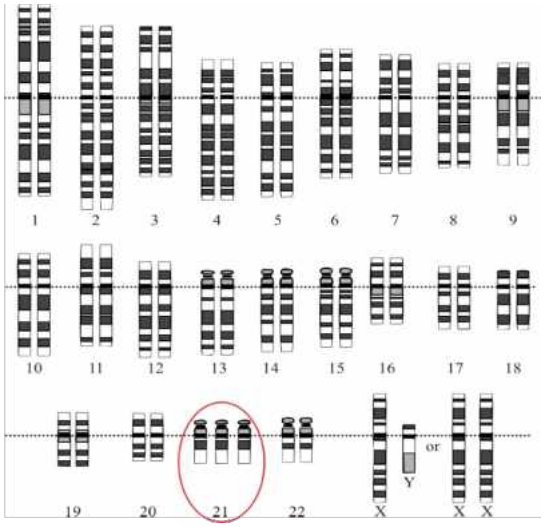


Figure 19.25: Karyotype of Down syndrome

Its symptoms include mental impairment, stunted growth, increased skin on back and neck, flat facial features, small head, short neck and limbs, upward slanting eyes, small ears, poor muscles, a protruding tongue, and heart defects. Males with Down syndrome are usually sterile, while females have lower rates of fertility relative to normal females.

Proper management of affected children can improve the quality of life of people with Down syndrome e.g., screening for common problems, medical treatment where indicated, a good family environment, and special education and proper care.

Tidbit

If an XX egg fuses with an X sperm, the female child will have 47 chromosomes with XXX sex chromosomes. This Trisomy in X is known as triple X syndrome. Such females show symptoms of learning disabilities, tall stature, and wide-spaced eyes. They are fertile and puberty starts at normal age.

2- Klinefelter Syndrome

If non-disjunction of X chromosome occurs during meiosis in mother, the egg may have both X chromosomes (XX) or may have no sex chromosome (designated as O). If an egg with XX chromosomes is fertilized by a normal sperm, the offspring will have **trisomy** (XXY) in sex chromosomes and the total number of chromosomes will be 47 (2n+1 or 44+XXY). Such non-disjunction may also occur in father during gamete

formation. It can result with a sperm having both X and Y chromosomes i.e., XY. If such sperm fertilizes a normal egg (X) it may result in XXY chromosomes in child. This condition (XXY) is known as Klinefelter syndrome (named after American physician Harry **Klinefelter** who first described it in 1942).

The affected children grow to sterile males with many female body characteristics (e.g., less body hair, breast growth), small testicles, and abnormal body proportions (long legs, short trunk, and shoulder equal to hip size). Their voices may not be as deep. The treatments include giving testosterone (to compensate testosterone deficiency at onset of puberty), and surgical removal of breast. Behavioural therapy is given to help in language disorders, difficulties at school, and socialization.

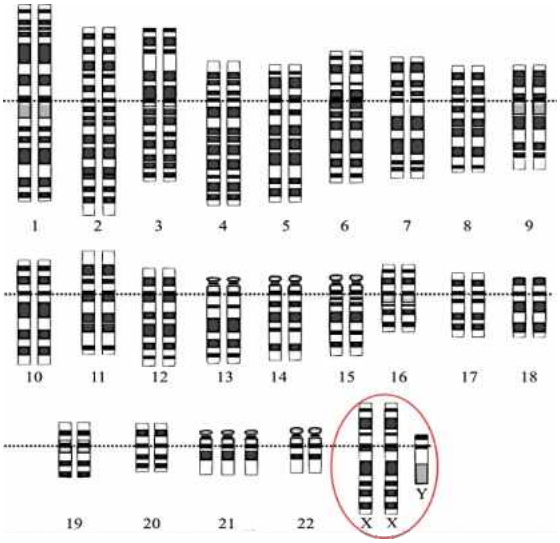


Figure 19.26: Klinefelter's syndrome karyotype

3- Turner Syndrome

If an egg with no X chromosome (O) fuses with a normal Y sperm, the resulting OY zygote fails to develop further because humans cannot survive without the genes on X chromosome. On the other hand, if an O egg fuses with a normal X sperm (or a O sperm fuses with a normal X egg), the zygote develops into female baby with 45 chromosomes (XO or 2n-1 or 44+XO) i.e., **monosomy** in sex chromosomes. The affected female is sterile and the condition is called Turner syndrome (American endocrinologist Dr. Henry **Turner** first described it in 1938).

The major symptoms include short stature, webbed neck, immature sex organs and infertility, low mental abilities, and small chin and jaw. Turner syndrome is also associated many health problems, such as liver and kidney issues, obesity, diabetes, and hypertension. Turner syndrome occurs roughly once in every 5000 female births.

Growth hormone treatment can improve growth. The affected female can have adult height, if growth hormone treatment is started early in childhood. Estrogen replacement therapy is used for the development of secondary sexual characteristics.

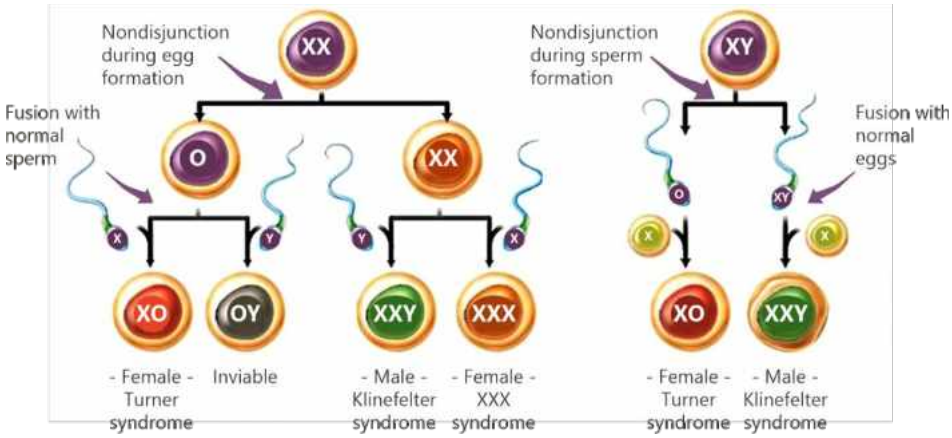


Figure 19.27: Nondisjunction of sex chromosomes

Tidbit

If a zygote receives one less autosome, it cannot complete development. Most of the zygotes with an extra autosome also do not survive. However, the chromosome pairs 13, 15, 18, 21, and 2 can have three chromosomes and the individual can survive for a time. The presence of an extra chromosome in pairs 13, 15, or 18 causes severe developmental defects, and infants with such a genetic makeup die within a few months.

In contrast, individuals who have an extra copy of chromosome 21 or 22, usually survive to adulthood. In such individuals, the maturation of the skeletal system is delayed, so they generally are short and have poor muscles. Their mental development is also affected.

EXERCISE

SECTION 1: MULTIPLE CHOICE QUESTIONS

16. Which of the following looks like beads on a string?
    (a) Chromosomes (b) Chromatin
    (c) Nucleosomes (d) Heterochromatin
17. The most convincing proof that DNA is the genetic material was provided by;
    (a) Mendel (b) Morgan
    (c) Hershey & Chase (d) Meselson and Stahl
18. Degeneracy of genetic code means that;
    (a) A codon consists of 3 nucleotides
    (b) Multiple codons can specify the same amino acid
    (c) Same codon can specify multiple amino acids
    (d) Each codon is read independently of the adjacent codons

19. How many codons are needed to specify for five amino acids?
(a) 3 (b) 5 (c) 15 (d) 30

20. Which of the following is NOT involved in the initiation of replication?
(a) DNA ligase (b) DNA gyrase
(c) single-stranded binding protein (d) Primase

21. Why DNA polymerase synthesises the leading and lagging strands of DNA in different manners?
(a) The origins of replication occur only at the 5′ end
(b) DNA ligase works only in the 3′ to 5′ direction
(c) DNA polymerase can only work on one strand at a time
(d) DNA polymerase can join new nucleotides only to the 3′ end of strand

22. Synthesis of a new DNA strand usually begins with:
(a) RNA primer (b) DNA primer
(c) DNA ligase (d) Okazaki fragment

23. The elongation of the leading strand during DNA synthesis;
(a) Progresses away from the replication fork
(b) Occurs in the 3′ – 5′ direction
(c) Produces Okazaki fragments
(d) Depends on the action of polymerase

24. Which of the following enzymes remove supercoiling in replicating DNA ahead of the replication fork?
(a) DNA polymerases (b) Helicases
(c) Primases (d) Topoisomerases

25. Which enzyme joins the bits of DNA?
(a) DNA polymerase (b) DNA ligase
(c) Endonuclease (d) Primase

26. Which of these subunits of RNA polymerase is required to initiate transcription?
(a) Alpha (α) (b) Sigma (σ)
(c) Omega (ω) (d) Beta (β)

27. When and where does the splicing of mRNA occur?
(a) Before transcription in nucleus (b) After transcription in nucleus
(c) After transcription in cytoplasm (d)After mRNA attachment with ribosome

28. What happens in mRNA splicing?
(a) Introns and exons are removed from the gene
(b) Introns and exons are joined together
(c) Exons are removed and introns are joined together
(d) Introns are removed and exons are joined together

29. Which of these happens first during protein synthesis?
    (a) Peptide bond formation
    (b) Binding of aminoacyl t-RNA with ribosome
    (c) Translocation
    (d) Binding of release factor with ribosome
30. The synthesis of protein chains is initiated with the amino acid;
    (a) Arginine (b) Methionine (c) Serine (d) Valine
31. The main function of tRNA is to:
    (a) Bind the mRNA with the smaller ribosomal subunit
    (b) Form a portion of ribosomes
    (c) Bind the complementary RNA and DNA strands together
    (d) bring amino acids from the cytoplasm to the ribosomes?
32. Ultraviolet radiation can cause mutations by;
    (a) Making double covalent bonds between adjacent pyrimidine bases
    (b) Breaking phosphodiester bonds in DNA strands
    (c) Adding nucleotides in NA
    (d) Substituting nucleotides in DNA
33. Phenylketonuria is a genetic disorder with a defect in the metabolism of:
    (a) Vitamin (b) Glucose (c) Amino acid (d) Lipid
34. The condition sickle cell anaemia is due to;
    (a) Deletion type point mutation
    (b) Substitution type point mutation
    (c) Insertion type point mutation
    (d) Chromosomal rearrangement

SECTION 2: SHORT QUESTIONS

17. What are the four types of chromosomes on the basis of position of centromere?
18. What is chromatin fibre? How is it formed during condensation of chromatin?
19. Briefly describe the concept of gene and gene locus.
20. State the functions of DNA polymerase I, II and III.
21. State the central dogma of gene expression.
22. What do you mean by "degeneracy of genetic code"?
23. Enlist some commonly occurring minor mutations in human.
24. Differentiate between:
    - Histone and nucleosome
    - Heterochromatin and euchromatin
    - Conservative and semi-conservative model of DNA replication
    - DNA helicase and DNA gyrase
    - DNA polymerase I and II

- Leading strand and lagging strand of DNA
- Translation and transcription
- Genetic code and codon
- Codon and anticodon
- Gene and allele
- Intron and exon
- Start codon and stop codon
- Non-sense codon and sense codon
- Repressor and activator proteins
- Point mutation and chromosomal mutation
- Down syndrome and Klinefelter syndrome
- Klinefelter syndrome and Tuners syndrome

SECTION 3: LONG QUESTIONS

15. Annotate the detailed structure of a chromosome.
16. Explain the concept of alleles as the alternative forms of a gene.
17. Narrate the experimental work of Griffith that proved that DNA is the hereditary material.
18. Narrate the experimental work of Hershey and Chase that proved that DNA is the hereditary material.
19. Describe the three models proposed about the mechanism of DNA replication.
20. Narrate the work of Meselson and Stahl to justify the semi-conservative replication.
21. Describe the events of the process of DNA replication.
22. Explain the mechanism of transcription.
23. Describe the mechanism of protein synthesis.
24. Describe the difference between protein synthesis in prokaryotes and eukaryotes.
25. Describe the importance of the regulation of gene expression.
26. Describe the negative and positive control of gene expression.
27. Explain the post-transcriptional modification of mRNA.
28. Write a note on point mutations and their sources.
29. Describe the symptoms, causes and treatments Down syndrome and Klinefelter syndrome.
30. Describe the symptoms, causes and treatments Turner syndrome.
31. Describe the symptoms, causes and treatments of Sickle cell anaemia and phenylketonuria.

INQUISITIVE QUESTIONS

7. DNA is tightly wrapped around histone proteins. How can replication enzymes read the code if it is buried inside a coil?

8. Our genetic code uses three-letter "words" to create 20 amino acids. Could a complex human exist if the code used only two-letter words?
9. DNA Polymerase III is a highly efficient proof-reader that fixes its own mistakes. What happens to a cell if the gene for the "proof-reader" enzyme itself gets a mutation?
10. Why the length of transcribed mRNA molecule (in eukaryotes) shortens as it enters the cytoplasm for translation?
11. How many types of tRNA molecules are necessary for a living cell, if the genetic code is a triplet code?
12. After isotopic labelling of the starting DNA with15N, Meselson and Stahl continue their experiment to at least two rounds of replication instead of stopping the experiment after only one round of replication. Justify their decision to continue to at least two replications.

Chapter 20

BIOTECHNOLOGY

STUDENTS'LEARNING OUTCOMES

After studying this chapter, the students will be able to:

- Introduce genetic engineering.
- Outline the function of restriction enzymes.
- Describe plasmids as vectors and explain how recombinant plasmids can be formed.
- Explain polymerase chain reaction (PCR).
- Define genetically modified organism.
- Explain the formation of human insulin protein in bacteria.

The word "Biotechnology" means "technology based on biology". In scientific terms, it is defined as the use of organisms, biological processes, or systems to make products that improve human lives and environment. Its main focus is to solve complex challenges in medicine, agriculture, and industry. Biotechnology is a vast field but this chapter would deal with the most important component of biotechnology i.e., genetic engineering.

For information

Traditional biotechnology has been used for thousands of years in the form of fermentation to create food products like bread, cheese, and beer. Modern biotechnology emerged in the 20th century, after the discovery of the structure of DNA and advanced molecular techniques.

20.1- Genetic Engineering

Biotechnology is a broad "umbrella" term. However, the true "engine" of modern biotechnology is "Genetic Engineering". Genetic engineering is defined as the alteration of an organism's DNA to change its characters (traits) or capabilities. It involves inserting, deleting, or modifying genes, and sometimes transferring genes between different species. In genetic engineering, scientists rely on two basic technologies:

1. Recombinant DNA Technology: Physically cutting a piece of DNA from one organism and "pasting" it into the DNA of another.
2. Polymerase Chain Reaction (PCR): Rapidly amplifying a tiny segment of DNA into a large quantity.

20.2- Recombinant DNA Technology

Recombinant DNA technology means the cutting, splicing, and combining DNA molecules from different sources into a single hybrid molecule. It also involves the

introduction of recombinant DNA (rDNA) into a host (e.g., bacteria) to produce new genetic combinations or specific proteins.

20.2.1- Tools used in Recombinant DNA Technology

1- Gene of Interest

The gene of interest is the gene which is to be manipulated. The gene of interest can be obtained by one of the following three ways:

i. Direct **synthesis** of the gene of interest in the lab by using different enzymes (e.g., polymerases and ligases).
ii. Synthesis by doing **reverse transcription** of its mRNA by using reverse transcriptase enzymes.
iii. **Cleavage from chromosome** by using enzymes restriction endonucleases.

2- Restriction Endonucleases

These enzymes cut specific sequences of nucleotides in DNA. They are also called molecular scissors. There are many kinds of restriction endonuclease. Each kind recognizes a specific nucleotide sequence in a DNA. Such a sequence of nucleotides is called **restriction site** or recognition region. The enzyme binds to the DNA at that sequence, and cuts the DNA at a particular place within this site.

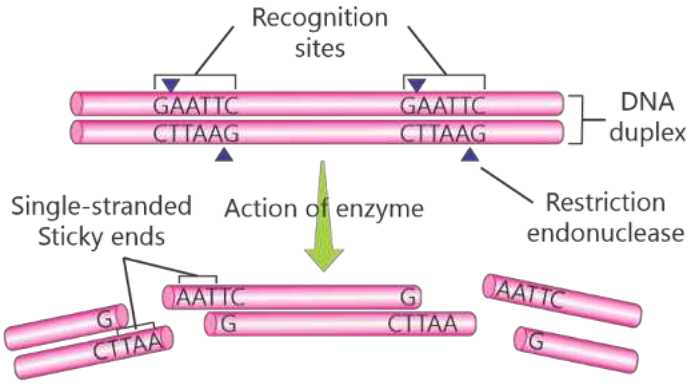


Figure 20.1: Action of restriction endonuclease

For information
Restriction endonucleases are valuable due to the fact that any two DNA fragments produced by the same restriction endonuclease can be joined together.

The restriction sites have **palindromic sequences** of four to eight nucleotides. This means the nucleotides at one end of the restriction site are complementary to those at the other end. So, the two strands of the DNA have the same nucleotide sequence running in opposite directions at the restriction site. Restriction endonuclease binds to and cleaves both strands. After cleavage, both DNA fragments have single-stranded ends (a few nucleotides long) that are sticky i.e., complementary to each other.

3- Vectors

Special molecules called vectors (or molecular carrier) are used for the introduction the of gene of interest into host cells. The gene of interest is attached to the vector and the vectors becomes a **recombinant DNA** (rDNA). Then, the rDNA is introduced into the host cells. The **plasmids** of bacteria are the most commonly used. Other vectors include;

i- Bacteriophages (e.g., phage lambda, bacteriophage P1)
ii- Cosmids (plasmids that contain a small region of bacteriophage DNA called the cos sequence)
iii- Bacterial artificial chromosomes – BACs
iv- Yeast artificial chromosomes – YACs.
v- P1 artificial chromosomes (have features of both P1 vectors and Bacterial Artificial Chromosomes)

Animal viruses (e.g., human cold virus) also serve as vectors to carry genes of interest into monkey and human cells.

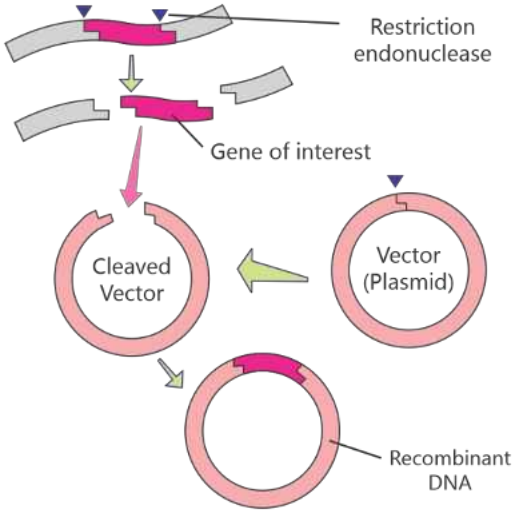


Figure 20.2: Use of Vector

4- DNA Ligase

This enzyme joins two DNA fragments. Therefore, it is called molecular glue. It forms the phosphodiester linkage between two adjacent nucleotides.

5- Expression System

Expression system means a suitable organism that can act as host for rDNA (gene of interest attached with vector) and can express the gene of interest. The selection of suitable expression system depends upon the type of vector used to make rDNA. Bacterial cells act as an ideal expression system because of their short generation time and simple genetic system.

20.2.2- Steps of Recombinant DNA Technology

1- Formation of Recombinant Vector (Plasmid)

i: DNA Cleavage: The source DNA of the organisms is **cleaved** by using a restriction endonuclease and the gene of interest is isolated. Similarly, the DNA of vector (e.g., plasmid or viral DNA) is also cleaved by using the same endonuclease. So, compatible sticky ends are produced at the gene of interest and the cleaved vector.

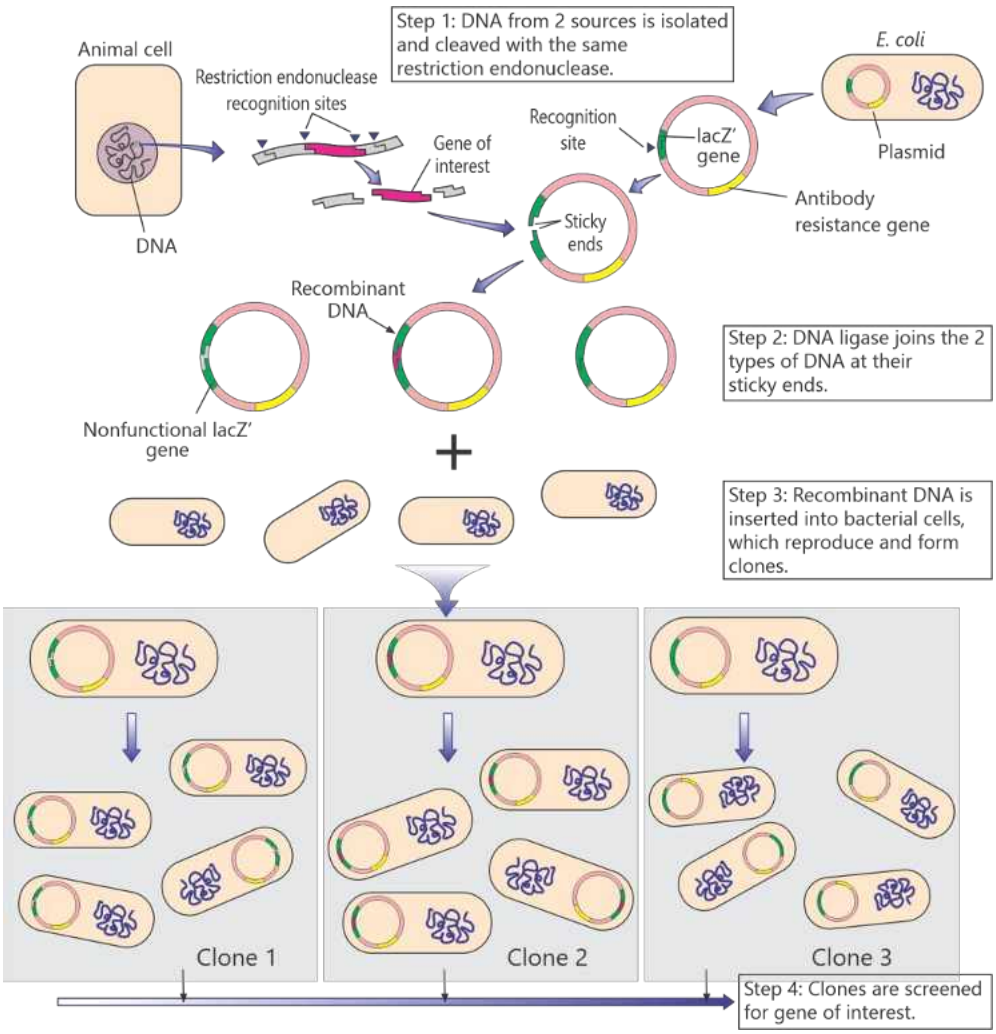


Figure 20.3: Steps of making Recombinant Vectors

ii: Production of Recombinant DNA (Vector): The gene of interest is inserted into the cleaved vector. For this purpose, the cleaved vector and the gene of interest are incubated together in the presence of DNA ligase. This enzyme connects them by forming phosphodiester linkages. So, the vector turns into recombinant vector (i.e., vector having the gene of interest).

2- Cloning of Recombinant Vector

The recombinant DNA (vector) and the expression system (usually bacteria) are incubated in the same medium. The bacterial cells take up recombinant DNA. All the incubated bacteria make bacterial clones which also contain the identical copies i.e., clones of the gene of interest.

3- Screening of Clone Library

Once the vectors have been inserted into host cells, scientists identify which bacteria actually took up the recombinant plasmid. This is done through:

i: Selecting for the Vector (Antibiotic Resistance): The plasmid contains an antibiotic resistance gene. The bacteria are grown on a medium containing that antibiotic (e.g., ampicillin). Only cells that took up a plasmid will survive and grow.

ii: Selecting for the Gene of Interest (Blue-White Screening): To distinguish between a "plain" plasmid and a "recombinant plasmid", the **lacZ' gene** of the plasmid is used. The lacZ' gene produces an enzyme which metabolizes a specific sugar and turns it blue

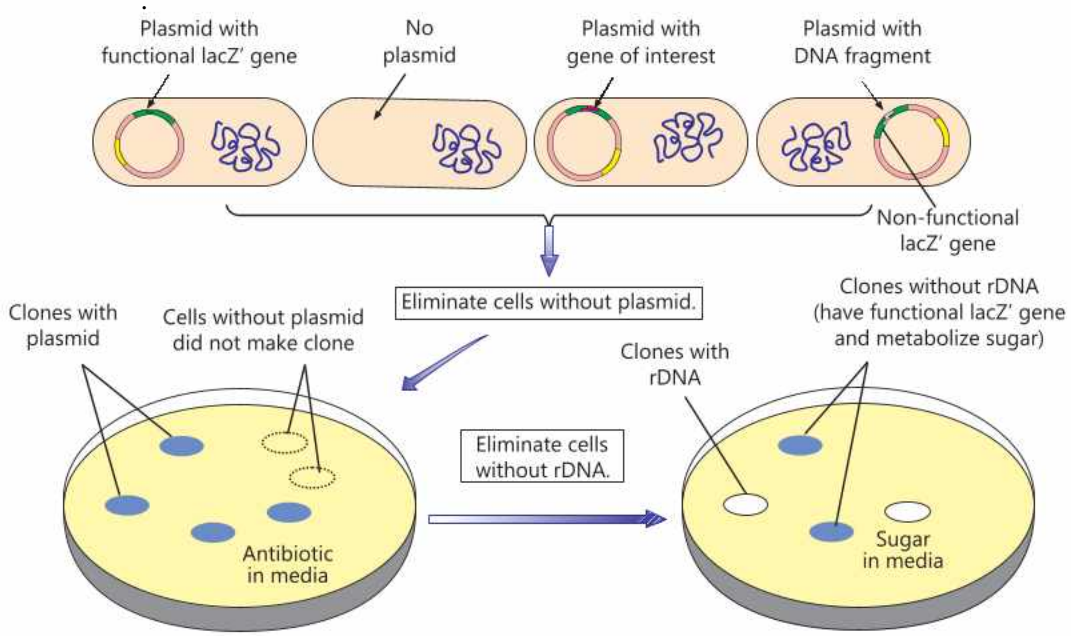


Figure 20.4: Screening of clones (Elimination of clones without vector (plasmid) and elimination of clones with vector (plasmid) but without recombinant DNA)

If the plasmid is not recombinant, the lacZ' gene remains functional and turns the bacterial colony blue. If gene of interest was successfully inserted, it "breaks" the lacZ' gene. Therefore, the desired recombinant clones will remain white.

iii: Identifying the Specific Gene (DNA Hybridization): To confirm the presence of gene of interest, a radioactive probe is used. It is a DNA strand complementary to gene of interest. The probe sticks (hybridizes) only to the colonies containing the gene of interest. Auto-radiography (photographic film) is used to locate these specific colonies on the master plate. The main steps of this technique are mentioned in the following diagram.

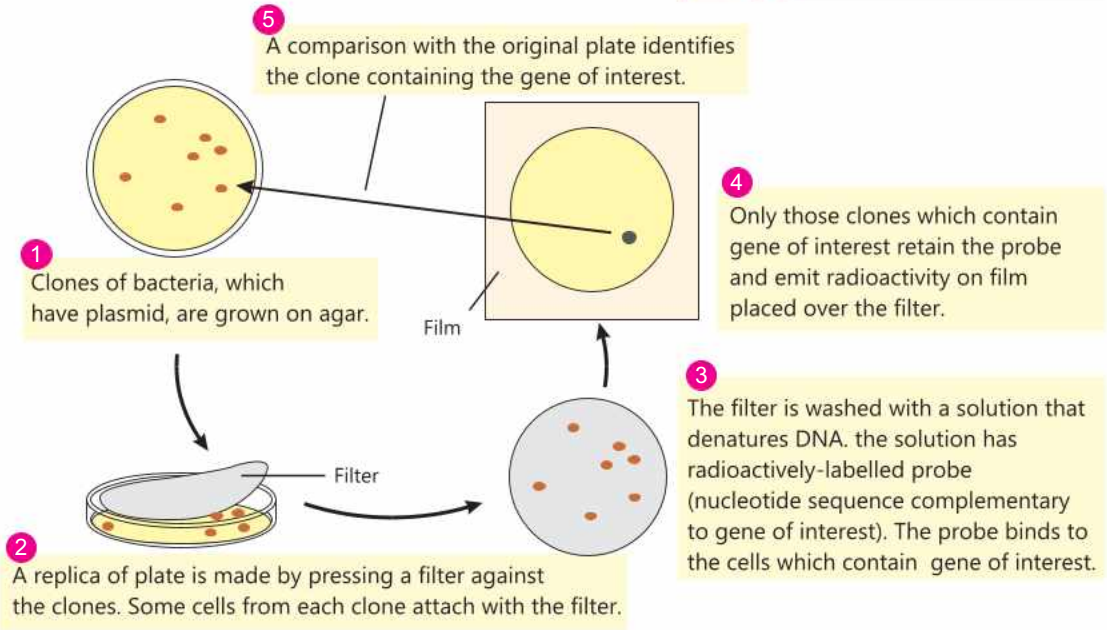


Figure 20.5: Identification of the clones that have gene of interest

20.3- Polymerase Chain Reaction (PCR)

In this technique, multiple identical copies of the gene of interest are made. The enzyme DNA polymerase is used to copy which polymerizes a given piece of DNA again and again, so that multiple copies are produced. That's why, the technique is known as polymerase chain reaction (PCR). Kary Mullis developed PCR in 1983. He won the Nobel Prize in Chemistry in 1993.

20.3.1- Steps of PCR

Step 1: Denaturation

First, an excessive amount of primer (a sequence of 20 to 30 nucleotides) is mixed with the DNA fragment (gene of interest). This mixture of primers and DNA is heated to about 98°C. At this temperature, the double-stranded DNA breaks into single strands. Each single stranded DNA can act as the template for the in vitro DNA synthesis.

Step 2: Annealing of Primer

The solution is allowed to cool to about 60°C. As it cools, the forward and the backward primers anneal or hybridize to the single-stranded DNA at its complementary regions. So, the rest of the fragment remains single-stranded.

Step 3: Extension of Primer

A very heat-stable DNA polymerase, called **Taq polymerase** is added.

All four types of nucleotides are also added. The Taq polymerase adds new nucleotides to the 3′ end of primer. In this way, it copies the rest of the fragment just like the replication of DNA. In this way, a complimentary copy of the entire single-stranded DNA has been added to the primer. Because both DNA strands are replicated, there are now two copies of the original DNA fragment.

Step 4: Repeating the Cycle

Steps 1 to 3 are repeated and the two copies become four. It is not necessary to add any more polymerase, as the heating does not harm this enzyme. Each cycle is completed in one or two minutes and it doubles the number of DNA molecules. After 20 cycles, a single fragment of DNA produces more than one million copies.

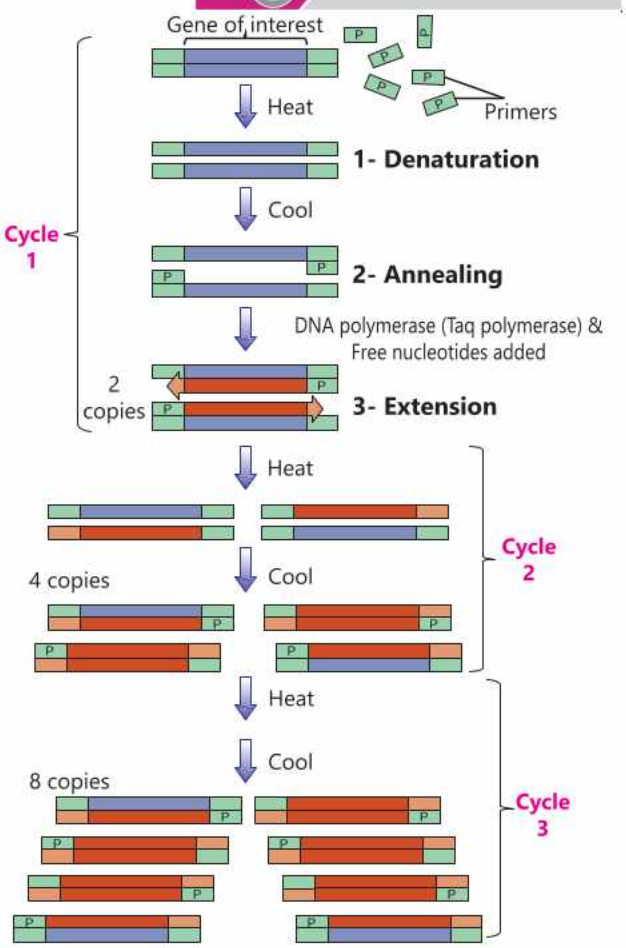


Figure 20.6: Steps of PCR

For information

PCR is also used to build genomic library. It is the collection of the total DNA from a single organism. Genomic libraries are commonly used for sequencing the DNA.

For information

The Taq polymerase is a temperature-tolerant enzyme isolated from *Thermus aquaticus*, a bacterium found in hot springs. This enzyme is stable and active at near-boiling temperatures.

For information

The optimum temperature for carrying out the extension of primer is 72°C. This step is completed in just one minute.

Applications of PCR

i. It is an efficient diagnostic technique used for detecting genotypes of infectious agents.
ii. It is used to detect genetic mutations responsible for certain genetic diseases and cancers.
iii. In criminal investigations, "DNA fingerprints" are prepared from the cells in a tiny sample of dried blood or at the base of a single human hair.
iv. Physicians can detect genetic defects in very early embryos by collecting a few sloughed-off cells and amplifying their DNA.
v. PCR is also used to examine the DNA of extinct species, as long as even a minuscule amount of their DNA remains intact.

20.4- Genetically Modified Organism (GMO)

A Genetically Modified Organism is a plant, animal or microbe in which the genome has been changed. Transgenic organisms are a specific subset of GMOs that have received DNA from a different, unrelated species. Approximately 90% of canola, cotton, corn, soybean, and sugar beets grown in the world are transgenic.

Methods of Gene Transfer

The gene (DNA) is transferred into the target cell through cell membrane or cell wall without destroying the cell. To produce a whole multicellular GMO, the gene is transferred into a gamete or gamete-producing tissue. Such gametes are then fused to produce GMOs. The following are common techniques used to transfer the gene of interest into a target cell.

(a) In **electroporation**, pores are created in the cell wall or cell membrane of target cell. The pores allow gene intake. Once the electrical current is discharged, the pores are closed.
(b) In **microinjection**, a fine needle, or injection, is used to directly inject the gene into the target cell nucleus.
(c) In **biolistic transformation**, the gene of interest coated with heavy metal ions is inserted into the target cell by mechanical force e.g., gene gun.
(d) In **protoplast mediated technique**, the cell wall and cell membrane are removed from the target cell. This protoplast is mixed with the medium containing the gene of interest.
(e) In **chemical methods**, the naked DNA is fused with a chemical agent. The chemical and DNA complex enters into the cell cytoplasm through diffusion or endocytosis.
(f) In **virus mediated gene transfer**, Adenovirus, Adeno-associated virus, HIV, lentivirus and other retroviruses are used for gene transfer.

20.4.1- GMO (Transgenic) Bacteria

Genes from a wide range of organisms can be added to a plasmid and inserted into bacteria. Transgenic bacteria can synthesize large amounts of the proteins encoded by the inserted foreign genes. Different forms of transgenic bacteria are being used to;

1. Produce pharmaceutical products e.g., insulin (to treat diabetes), clotting factors (to treat haemophilia), hepatitis B vaccine (against hepatitis B), Covid-19 vaccine (against Corona), human growth hormone (to treat dwarfism), interferon (to treat some cancers), erythropoietin (to treat anaemia), and tissue plasminogen activator (which dissolves blood clots).
2. Produce many food products e.g., alpha-amylase (converts starch to simple sugars), chymosin (clots milk protein for cheese making), and pectin-esterase (improves fruit juice clarity).
3. Produce enzymes which degrade toxins to convert pollutants into a less toxic form.
4. Leach copper from ore, clean up mercury pollution and detect arsenic in drinking water.

20.4.2- GMO (Transgenic) Animals

A transgenic animal carries a foreign gene to produce the desired protein. Transgenic animals serve various purposes e.g., producing drugs, enhancing yields, increasing resistance to disease etc. For example;

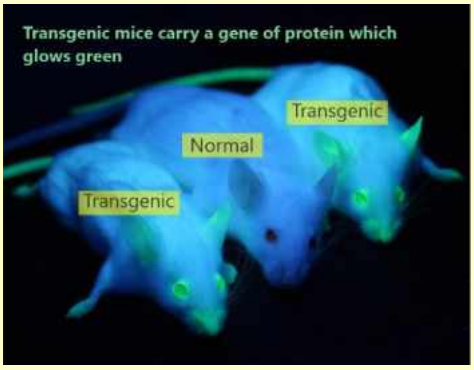
1. Transgenic dairy cows have genes from human beings to produce milk that is the same as human breast milk.

Tidbit

In theory, foreign genes may be inserted either in germline or in somatic cells of humans. If genes are inserted in germline cells, it results in inheritable changes. But, most research has focused on somatic cells because of risks and ethical issues associated with germline transformation.

Tidbit

Since the mid-1980s, transgenic mice are a key model for investigating diseases. Mice are the model for this purpose because there is complete analysis of its genome sequence and its genome is very much similar to humans.



2. Transgenic goats produce milk with strong spiderweb-like silk proteins.

3. Transgenic goats also produce human enzyme lysozyme in their milk to fight diarrhoea causing bacteria in humans.
4. Transgenic fish show better growth, reduced food intake, increased cold tolerance, and disease resistance.
5. In gene therapy, genetically modified viruses are used to deliver genes to humans to cure various inheritable diseases such as abnormal development of T and B cells and inherited blindness.
6. The first human biological drug produced from a transgenic goat was an anticoagulant to reduce the probability of blood clots during surgery or childbirth.
7. Malaria-resistant mosquitoes have been developed in the laboratory by inserting a gene that reduces the development of *Plasmodium* in them.

20.4.3- Transgenic Plants

Many plant cells are totipotent, meaning that a single cell can form a new plant. Biologists grow a new plant from a transgenic cell of plant. The main purposes of producing transgenic plants are;

1. Genes of *Bacillus thuringiensis* (a soil bacterium) have been inserted in many plants (e.g., cotton, rice, maize, potato, tomato, brinjal, cauliflower, cabbage etc.). These genes make proteins that kill insect pests but do not harm other useful insects.

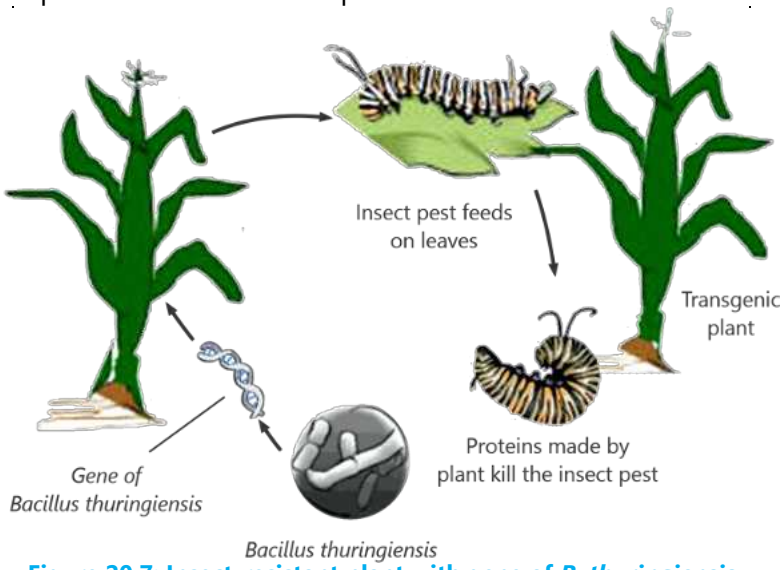


Figure 20.7: Insect-resistant plant with gene of *B. thuringiensis*

For Information

Each year, deficiency of iron affects 1.4 billion women and deficiency of vitamin A kills 670,000 children and causes 500,000 cases of irreversible childhood blindness.

2. Many crops have been modified to be resistant against glyphosate (an herbicide).

3. In developing countries, biologists have made GMO (transgenic) rice (golden rice) in which they have inserted genes to increase iron content. These genes were taken from beans, *Aspergillus fungus*, and wild rice. Similarly, the genes for making beta-carotene (a precursor of vitamin A) were taken from daffodil and inserted in rice. The transgenic rice is grown and consumed in areas with a shortage of dietary iron and vitamin A
4. GMO plants and plant cells are used for the production of biopharmaceuticals e.g., cytokines, hormones, antibodies, enzymes and vaccines etc.

20.5- Formation of Human Insulin in Bacteria

The production of human insulin was the first major triumph of genetic engineers. By using bacteria, scientists produce vast quantities of insulin that is identical to the insulin hormone naturally made in the pancreas. The process of creating synthetic human insulin involves producing the two polypeptide chains separately and then joining them chemically.

1. Preparation of the Two Genes: Since insulin consists of two distinct chains, scientists synthesise two different DNA sequences: one for Chain A and one for Chain B.

2. Construction of Recombinant Plasmids: Two sets of plasmids are obtained. The DNA for Chain A is inserted into one set of plasmids, and the DNA for Chain B is inserted into another. In both cases, the insulin gene is often attached next to a bacterial gene (like lacZ) to help the bacteria "recognize" and express the foreign DNA.

3. Transformation of Host Cells: The recombinant plasmids are introduced into two separate cultures of *Escherichia coli* bacteria. Culture 1 acts as a factory for Chain A. Culture 2 acts as a factory for Chain B.

4. Fermentation and Protein Expression: The bacteria are grown in large industrial bioreactors. As the bacteria multiply and perform their normal metabolic functions, they simultaneously "read" the human DNA and produce large quantities of the individual insulin chains.

5. Extraction and Chemical Joining: Once the chains are synthesized, the bacteria are harvested and the insulin chains are extracted and purified. In a laboratory setting, Chain A and Chain B are mixed together. They are linked by creating di-sulfide bonds (sulfur-to-sulfur bridges) between specific amino acids.

This final step creates the functional, three-dimensional human insulin molecule, identical to that produced by a healthy human pancreas.

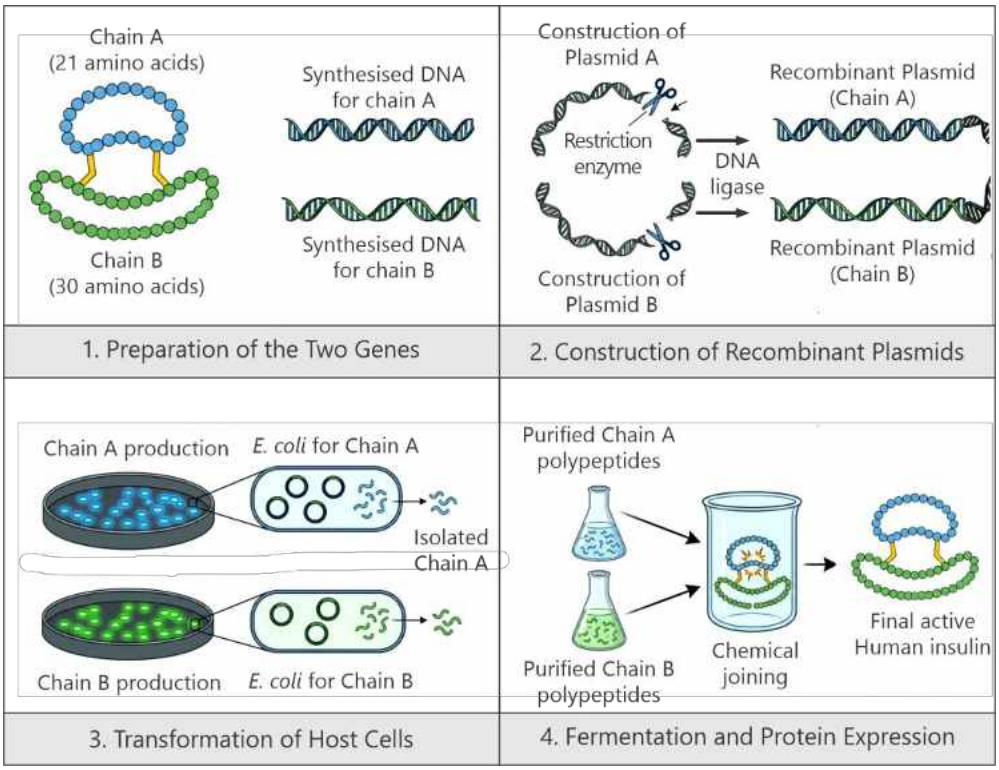


Figure 20.8: Formation of human insulin in bacteria

EXERCISE

SECTION 1: MULTIPLE CHOICE QUESTIONS

1. Identify the FALSE statement.
   (a) Restriction enzyme is used for the production of RFLPs.
   (b) DNA ligase enzyme is used to cut DNA, creating the sticky ends.
   (c) Reverse transcriptase enzyme is used for making cDNA from mRNA.
   (d) Electrophoresis means the separation of DNA fragments.
2. Which bacterium is used in the production of insulin by genetic engineering?
   (a) *Saccharomyces* b) *Rhizobium*
   (c) *Escherichia* (d) *Mycobacterium*
3. Which technique is used for cloning of gene/DNA?
   (a) RFLP analysis
   (b) Polymerase Chain Reaction (PCR)
   (c) Electroporation
   (d) Gel electrophoresis

4. Which method is used for obtaining the gene of interest?
   (a) Artificial gene synthesis (b) Reverse transcription
   (c) Cleavage from chromosome (d) All of these
5. When a gene of interest is attached to the vector, the new molecule is called;
   (a) Recombinant DNA
   (b) Restriction length fragment
   (c) Complimentary DNA
   (d) Genomic library
6. What is the use of plasmid in genetic engineering?
   (a) To carry DNA to a cell (b) To cut DNA
   (c) To join pieces of DNA (d) To replicate DNA
7. Which of these can serve as ideal expression system for gene cloning?
   (a) Bacteriophages (b) Bacterial cells
   (c) Plant cells (d) Animal cells
8. In PCR, optimum temperature for carrying out the extension of primer is;
   (a) 98° C. (b) 72° C. (c) 60° C (d) 37° C.
9. Which enzyme is used for the extension of primer in PCR?
   (a) DNA polymerase (b) Transcriptase
   (c) DNA ligase (d) Restriction endonuclease
10. What is true for a Genetically Modified Organism
   (a) Genes have been introduced (b) Genes have been enhanced
   (c) Genes have been deleted (d) All of these

SECTION 2: SHORT QUESTIONS

25. What is the role of restriction endonucleases and DNA ligases in gene cloning?
26. What is the role of vectors in recombinant DNA technology?
27. Describe how the gene of interest is selected and isolated during recombinant DNA technology.
28. State the steps for the integration of gene of interest into the vector.
29. State the role of GMO in making biotechnology products.
30. Differentiate between;
    - Endonuclease and ligase
    - PCR and Recombinant DNA Technology

SECTION 3: LONG QUESTIONS

32. Describe the components and steps of recombinant DNA technology.
33. Write a detailed note on restriction enzymes.
34. Write a note on polymerase chain reaction.
35. Describe the methods for the introduction of human DNA into bacteria.

INQUISITIVE QUESTIONS

13. Find and describe the accomplishments of the renowned genetic engineers working in private and public institutions in your province or city.
14. Analyse and interpret the DNA of a child by comparing it with that of two individuals in a case of disputed parenthood.
15. What may be the application of polymerase chain reaction?
16. The human genome project is regarded as the most ambitious project ever undertaken by man. Justify.
17. Investigate careers that require an understanding of biotechnology and genetic engineering.

Chapter 21

IMMUNITY

STUDENTS'LEARNING OUTCOMES

After studying this chapter, the students will be able to:

- List the structural features of human skin that make it an impenetrable barrier against invasion by microbes (1st line of defense).
- Explain how oil and sweat glands within the epidermis inhibit the growth and also kill microorganisms. (1st line of defense)
- Recognize the role of the acids of the digestive tract as killing bacteria present in food.
- State the role of the ciliated epithelium of the nasal cavity and the mucous of the bronchi and bronchioles in trapping airborne microorganisms.
- Describe the role of macrophages and neutrophils in killing bacteria.
- Explain how Natural Killer (NK) cells kill cells infected by microbes and cancer cells.
- State the way proteins of the complement system kill bacteria and that interferons inhibit viruses from infecting cells.
- State the events of the inflammatory response as a generalized, nonspecific defense.
- Outline the release of pyrogens by microbes and their effect on the hypothalamus to boost the body's temperature.
- List the ways that fever affects microbes.
- Define the specific immune system as providing specific defense and acting as the most powerful means of resisting infection.
- Identify monocytes, T- cells, and B-cells as components of the immune system.
- State inborn and acquired immunity as the two basic types of immunity.
- Differentiate between active and passive immunity as the two types of acquired immunity.
- Describe the role of T-cells in cell-mediated immunity.
- Describe the role of B-cells in antibody-mediated immunity.
- Discuss the role of T-cells and B-cells in transplant rejections.
- Evaluate the discovery of monoclonal antibodies and justify how this accomplishment revolutionized many aspects of biological research.
- Identify the process of vaccination as a means to develop active acquired immunity.
- Draw the structural model of an antibody molecule.
- Explain the role of memory cells in long- term immunity.
- Define allergies and correlate the symptoms of allergies with the release of histamines.
- Describe the autoimmune diseases with examples.

Most human diseases result from microbial infections – invasions of the body by viruses, pathogenic bacteria, fungi, or protists. To defend against infections and against cancers, vertebrates possess a defence system called the **immune system**. It continually checks the bloodstream for the presence of any foreign cells and molecules.

For example, when the immune system detects an infection, it attacks and destroys the invading microbes. That is why, the diseases that weaken the immune system, such as AIDS, are very dangerous.

Tidbit

Our body is defended from infection the same way that a city is defended against foreign invasion. There are walls to make invaders entry difficult, roaming policemen check strangers, security guards challenge anyone wandering and call policeman if a proper ID is not presented. Our body also has different lines of defence.

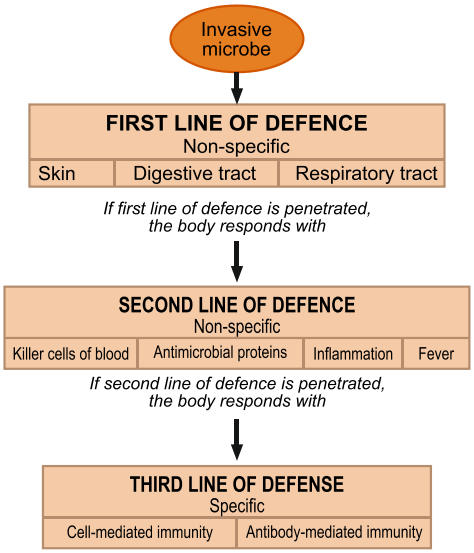


Figure 21.1: Lines of defence against infection

21.1- First Line Of Defence

The first line of defence against infections consists of skin, the digestive tract, and the respiratory tract.

21.1.1- Defence Provided by Skin

Skin is the outermost layer of our body. It provides the first defence against invasion of microbes. The epidermis of skin is 10 to 30 cells thick. The outer layer of epidermis is called **stratum corneum**. Its cells are continuously shed, because they are continuously damaged. New cells are formed in the deep epidermis from where they migrate upward and replace the worn-out cells. These cells form keratin protein which makes the skin tough and impenetrable for microbes.

For Information

Skin is the largest organ of the vertebrate body. In an adult human, 15% of the total weight is skin. One square centimetre of human skin contains 200 nerve endings, 10 hairs and muscles, 100 sweat glands, 15 oil glands, 3 blood vessels, 12 heat receptors, 2 cold receptors, and 25 pressure receptors.

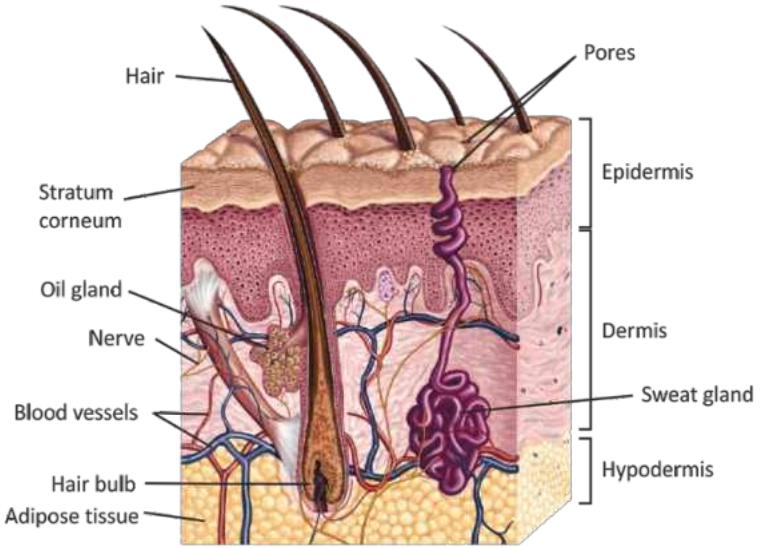


Figure 21.2: Layers of skin

Skin further reinforces the defence by secreting chemicals. The oil and sweat glands present in skin lower the pH at the skin's surface to 3-5. This is enough acid level that inhibits the growth of many microbes. Sweat also contains the enzyme lysozyme, which attacks and digests the cell walls of many bacteria.

21.1.2- Defence Provided by Digestive and Respiratory Tracts

Many bacteria are present in the food that humans eat. Most of these are killed by **saliva in the oral cavity**, which also contains lysozyme. The strong digestive acids present in the stomach and the protein-digesting enzymes in the intestine also kill bacteria.

Many microbes are present in the air that we breathe. Ciliated epithelial cells in the nasal cavity trap many bacteria before they can enter the air passageway. The cells lining the smaller bronchi and bronchioles secrete a layer of sticky **mucous** that traps microbes before they can reach the warm, moist lungs. Cilia on the cells lining these passages continually sweep the mucous upward, where it can be swallowed.

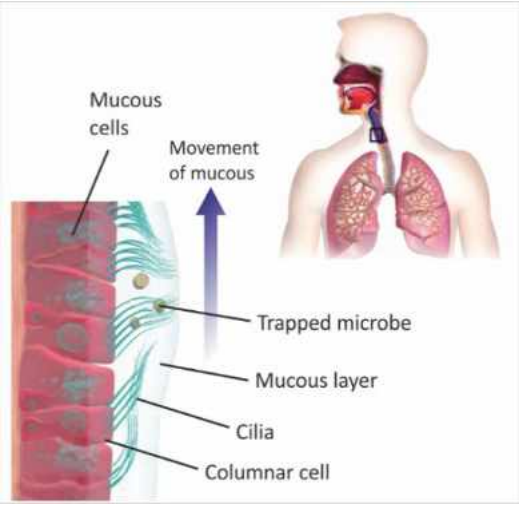


Figure 21.3: Epithelium of trachea, with trapped microbes

21.2- Second Line of Defence

The first line of defence of the body is very effective. However, it may occasionally be overcome by invaders. When such invaders reach deep tissues, a second line of defence comes into play. Along with the first line of defence, the second line is also categorized as a non-specific defence.

Human body uses a lot of non-specific cellular and chemical devices in its second line of defence. The four most important of these are; the germ-killer cells of blood, antimicrobial proteins, the inflammatory response, and the temperature response.

21.2.1- Germ-Killer Cells of Blood

The most important non-specific defence is provided by the germ-killer cell of the blood. They attack and kill the microbes. These cells patrol the bloodstream and wait for invaders. There are three basic kinds: macrophages, neutrophils, and natural killer cells.

1. Macrophages

Macrophages develop from monocytes (a type of white blood cells). A macrophage ingests a bacterium, by endocytosis, and kills it by its lysozyme enzymes. Some macrophages are fixed within some organs e.g., lungs, liver, spleen, and brain. But most of them patrol in the blood, lymph and interstitial fluid.

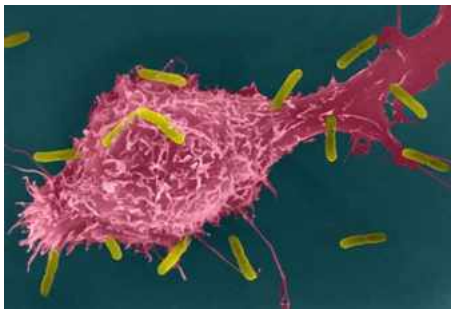


Figure 21.4: A macrophage attacking microbes

2. Neutrophils

Neutrophils are a type of white blood cells. They use three methods for killing pathogens. The first method is phagocytosis. In the second method, they release soluble anti-microbial chemicals on pathogens. In the third method, they generate and release network of DNA fibres. These fibres bind with pathogenic bacteria and kill them.

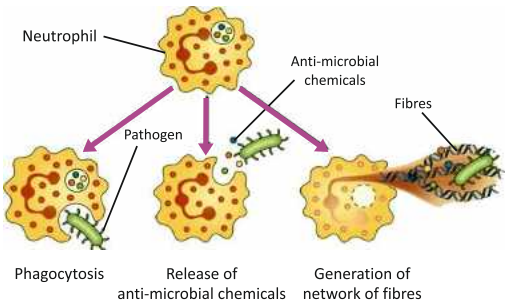


Figure 21.5: Methods of neutrophils to kill pathogens

3. Natural Killer Cells

These are a type of T-lymphocytes and are also called **cytotoxic T-cells**. They do not kill invading microbes, rather they detect and attack the body cells that have been infected with viruses. They release special proteins, called **perforins**, which insert into the membrane of the target cell. It results in the formation of a pore in the membrane of target cell. This pore allows water to rush into the target cell, which then swells and bursts. They are also able to detect cancer cells, which they kill before the cancer cells have a chance to develop into a tumour.

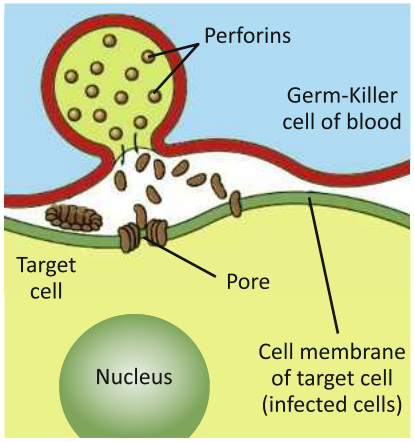


Figure 21.6: Natural killer cell attacking an infected cell

For Information

The patrolling cells (macrophages, neutrophils, and natural killer cells) do not attack their own body because all the normal cells of body contain a surface protein marker that identifies them. This protein is called self-protein. The germ-killer cells of blood do not attack the cells which have self-proteins. They only attack the cells that lacks such proteins.

21.2.2- Protective Proteins

Two main groups of proteins participate in the non-specific defence of the body.

1. Complement system proteins

Special proteins produced in the liver make the **complement system** of proteins. This system consists of more than a dozen different proteins that circulate in the blood in an inactive state. When they encounter a microbe, they become active in a sequence. The first protein activates the second and so on. The final five proteins form a **membrane-attack complex** (MAC) into the plasma membrane of microbe. This complex punctures the plasma membrane of microbe. So, fluids and salts move inside microbe resulting in its swelling and bursting.

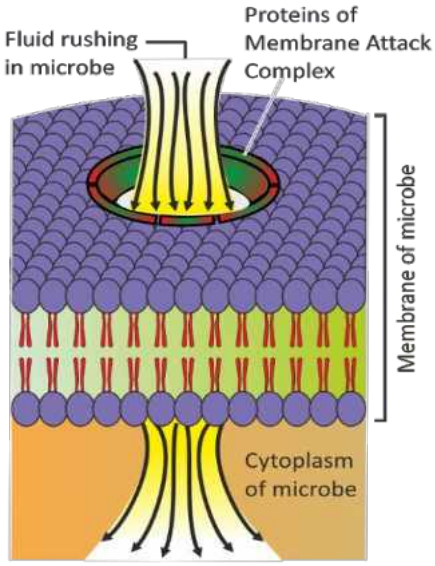


Figure 21.7: The Complement system of proteins and their functioning

2. Interferons

Interferons are another class of protective proteins that play a key role in body's defence. They are released by virus-infected cells. They cause nearby cells to enhance

their anti-viral defences. Interferons belong to the large class of proteins known as **cytokines**. These proteins work for communication between cells to trigger the protective defences of the immune system. Interferons also activate other immune cells, such as natural killer cells and macrophages.

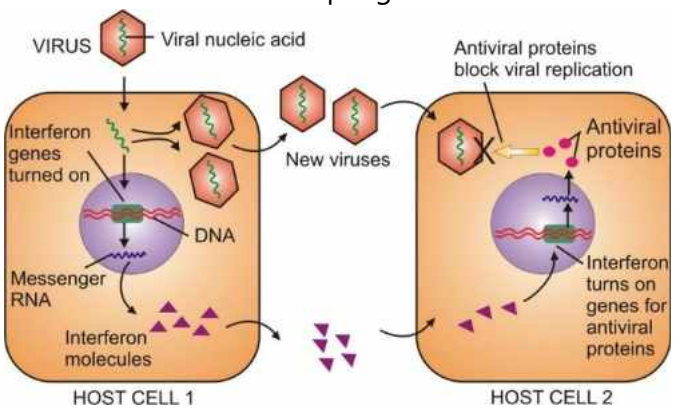


Figure 21.8: The interferon mechanism to alert other cells

21.2.3- Inflammatory Response

It is one of the most generalized non-specific responses to infections. Infected or injured cells release chemical signals such as **histamines** and **prostaglandins**. These chemicals cause local expansion of blood vessels. In this way, the capillary blood flow is increased at the site of injury or infection. The capillaries also become more permeable. It results in redness, swelling and warmth at the spot. This condition is called **inflammation**. The heat makes the site unfavourable for microbial growth. It also promotes healing and raises mobility of white blood cells. More phagocytes (macrophages and neutrophils) migrate from the blood to the inflamed tissue, where they engulf microbes. Monocytes arrive and release chemicals to kill the microbes and then macrophages clean up dead microbes, cells, and debris. The inflammatory response is often strong enough to stop the spread of microbes.

For Information

The pus associated with some infections is a mixture of dead or dying neutrophils, broken down tissue cells, and dead pathogens. In some cases (for example, arthritis) inflammation occurs in the absence of infection or injury. This is an example of misdirected immune response.

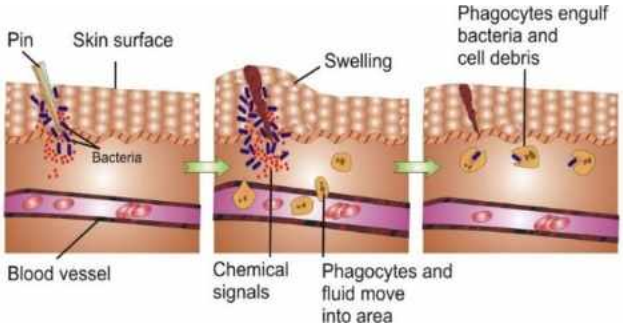


Figure 21.9: The inflammatory response

21.2.4- Temperature Response

When macrophages attack the invading microbes, they release chemicals called **pyrogens**. The pyrogens pass through the bloodstream to the brain and reach the hypothalamus. It is the body's thermostat. Here, the pyrogens stimulate the hypothalamus to boost the body's core temperature above the normal value of 37 °C (98.6 °F). It results in the rise in body temperature. The higher-than-normal temperature is known as **fever (pyrexia)**. Fever acts as a defence mechanism against microbes. It hinders the ability of bacteria and viruses to reproduce. Some microbes (e.g., *Streptococcus pneumonia*, *Influenza*) are temperature-sensitive and are killed at high fever. Fever also enhances the speed of the actions of neutrophils and macrophages. During fever, the body releases more cytokines (interferons).

For Information
In general, temperatures greater than 103 °F (39.4 °C) are considered dangerous, and those greater than 105 °F (40.5 °C) are often fatal.

21.3- Third Line of Defence

Sometimes, microbes overcome the first and second lines of defence. When this happens, they face a third line of defence. The third line of defence is provided by the **immune system**. It is the specific defence, which means that it targets specific germs and acts especially against them. The third line of defence (immune system) can remember previous invaders and responds quickly if they enter the body again.

Types of Immunity

Non-specific defence (first and second line of defence) is present at birth. So, it is also called **inborn** or **innate immunity**. While the specific defence is acquired during growth and it is called **acquired immunity**. The acquired immunity is further of two types i.e., active and passive. In **active immunity**, the body itself prepares antibodies against the antigens of microbes. While, in **passive immunity**, the individual is given antibodies to combat specific microbes. Passive immunity is short-lived.

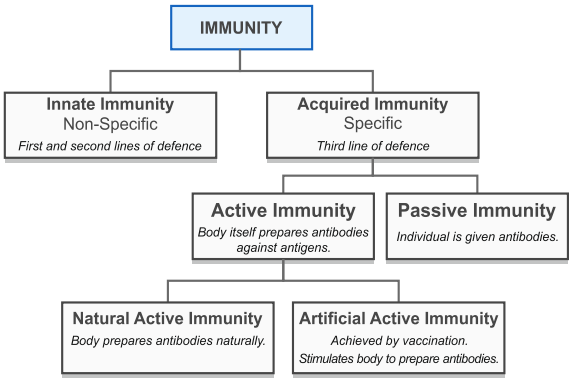


Figure 21.10: Types of immunity and their mechanisms

Active immunity may be natural or artificial. In **natural active immunity**, body prepares antibodies naturally when exposed to antigens. It is the most effective form of immunity. **Artificial active immunity** is achieved by administering weakened

antigens in the body (vaccination). In this case, the weak antigens stimulate the body to prepare antibodies against the real antigens.

21.3.1. Components of the Immune System

The immune system is not localized to one place in the body. Rather, it is composed of a lot of individual cells that rush to the infection site to combat invading microbes. These cells are the white blood cells which arise in the bone marrow and circulate in blood and lymph. They are also found in lymph nodes, spleen, liver, thymus, and bone marrow. There are three main kinds of white blood cells i.e., monocytes, T cells, and B cells. T and B cells are collectively called **lymphocytes**.

1. Monocytes

They are a type of WBCs and are produced in lymphoid tissues. They circulate in blood and also enter the interstitial fluid. Here, they mature into **macrophages**. The macrophages phagocytose and destroy the microbes. They also produce proteins that activate the helper T cells. Monocytes also mature into other kinds of phagocytes, which remain in liver, spleen, lymph nodes, and other tissues. They also help to clear cellular debris and assist in repairing tissue damage.

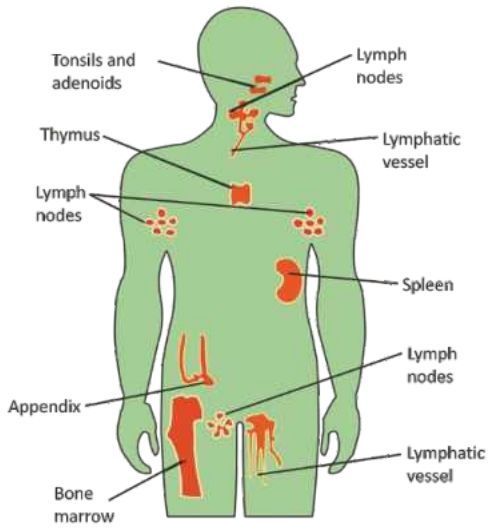


Figure 21.11: Major organs of the immune system

2. T Cells

Like other white blood cells, T cells also develop from the stem cells in the bone marrow. They migrate to the thymus, a small grey gland located just above the heart. There, they mature and develop the ability to identify foreign molecules (antigens) present on the invading microbes. Tens of millions of different T cells are made. Each T cell is specialized in recognizing one particular antigen. No invader can escape being recognized by at least a few T cells. There are four main kinds of T cells.

1. Helper T cells, which initiate the immune response
2. Cytotoxic T cells, which break the cells that have been infected by microbes
3. Inducer T cells, which supervise the development of T cells in the thymus
4. Suppressor T cells, which terminate the immune response

3. B Cells

The B cells also develop in bone marrow but unlike T cells, they do not travel to the thymus for maturity. They complete their maturation in the bone marrow. From

there they are released to circulate in the bloodstream and lymph. Individual B cells, like T cells, are specialized to recognize particular antigens. When a B cell encounters an antigen, it begins to divide into many new cells called **plasma cells**. Each plasma cell produces an antibody that sticks to the antigen, marking it for destruction.

21.3.2- Cell-Mediated and Antibody-Mediated Immunity

We have learned that when microbes attack body, macrophages provide the immediate response by engulfing them. This response cannot eliminate many microbes, but it gives time for the immune system (third line of defence) to respond.

When a macrophage engulfs a microbe, it also produces special proteins for the helper T cells. When the helper T cells recognize these proteins through their receptors, they are activated. The activated helper T cells initiate two parallel immune responses i.e., cell-mediated immunity, and antibody-mediated immunity.

1. Cell-mediated immunity

The cell-mediated immunity, carried out by T cells, protects the body from virus infections and cancer, killing abnormal or virus-infected cells. In cell-mediated immunity, cytotoxic T cells recognize and destroy infected body cells. Their mechanism of killing is the same as that of natural killer cells i.e., they puncture the membranes of the host target cells (infected cells or abnormal cancer cells). Important events in cell-mediated immunity are as follows.

1. **Proliferation:** When a helper T cell is activated, it produces soluble chemicals e.g., T cell growth factor. This factor starts the proliferation of all types of T cells and thus large clones of T cells are formed, capable of recognizing the antigens.
2. **Activation:** A second factor is secreted by the activated helper T cell, which attracts macrophages to the site of infection.
3. **Induction:** Helper T cells activate inducer T cells in the thymus, which trigger the maturation of lymphocytes into T cells.
4. **Attack:** The receptors present on the surface of cytotoxic T cells recognize the infected body cells and so these T cells bind with the infected cells. They disrupt the membrane of the infected cells, and thus the cells burst.
5. **Suppression:** Suppressor T cells block the response of cytotoxic T cells. The population of suppressor T cells multiplies more slowly than do the cytotoxic T cells. Their low initial numbers prevent them from blocking the cytotoxic attack. After 1 to 2 weeks, however, the number of suppressor T cells rises to the point where they are able to shut down the cytotoxic T cell response.
6. **Memory:** After suppression, a population of T cells persists, probably for the life of the individual. These helper and cytotoxic T cells are now called memory cells. And they provide a rapid response to any later encounter with the microbe again.

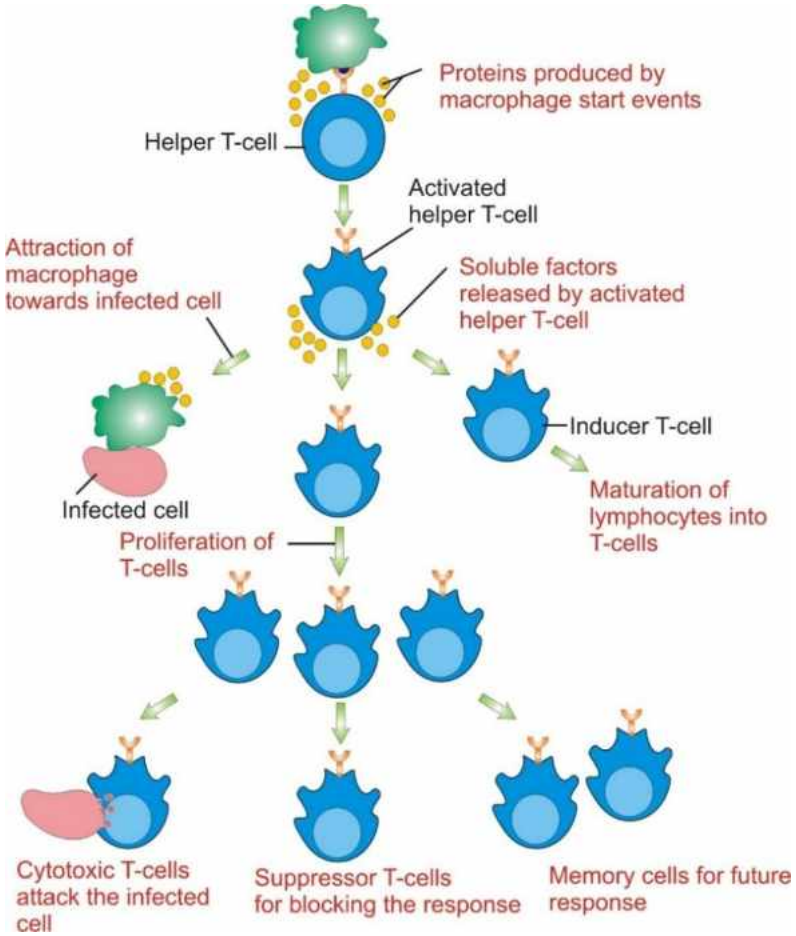


Figure 21.12: Events in cell-mediated immunity

2. Antibody-mediated immunity

When helper T cell is stimulated, it does not activate the cell-mediated immunity only. Raher, it also activates a second, stronger defence, called antibody-mediated or humoral immunity. The key players in this immunity are the B cells. They do not attack the antigens directly. Rather, they mark them for destruction. They also inactivate the toxic molecules. Important events in antibody-mediated immunity are as follows.

> Antibodies constitute about 20% by weight of the total protein in blood plasma.

Proliferation: Each B cell binds to the antigens through the B receptor proteins on its surface. Helper T cells recognize such antigen-bound B cells and bind with them. Then, the helper T cells release proteins that stimulate the B cell to proliferate. As a result, a large number of cells called **plasma cells** are produced from each B cell.

Differentiation and Secretion: Most plasma cells stop reproducing and start producing more B receptor proteins. The B receptor proteins are secreted and now called as antibodies. The plasma cells secrete a great number of antibodies in only a few days.

A vaccine stimulates the antibody production and formation of memory cells. Vaccines are made from killed or weakened pathogens that cause antibody production.

Attack: Antibodies do not destroy an antigen directly. Rather, they mark it for destruction by one of three mechanisms.

1. ***Complement:*** Some antibodies, when attached to an antigen, activate the proteins of the complement system. These proteins penetrate the membrane of the antibody-coated antigen and make holes in it. Water is drawn into the antigen cell, causing it to swell and burst.
2. ***Macrophages:*** Some antibodies, when attached to an antigen, are recognized by a macrophage, which ingests and breaks it.
3. ***Killer cells:*** Killer cells are similar to natural killer cells, except that these possess receptors that recognize antibody-coated antigens. When a killer cell encounters an antibody-coated antigen, it binds and kills the cell.

Immune System and Gene Therapy

Malignant melanoma is a lethal skin cancer. A special type of WBC called tumour-infiltrating lymphocyte (TIL) normally attacks a cancerous tumour. But, is not strong enough to control the tumour. Genetic engineers are trying to insert a gene into TIL. The gene will enable TIL to make a protein called tumour necrosis factor (TNF). This protein kills tumour cells. When genetically engineered TIL will be returned to patient's blood, they will enter a tumour and produce TNF to kill it.

Suppression: After several weeks, the suppressor T cells shut down the antibody response.

Memory: Some members of the clone of plasma cells do not produce antibodies. Instead, they persist as memory B cells. These cells provide a rapid **secondary response** to any later encounter with the same antigen.

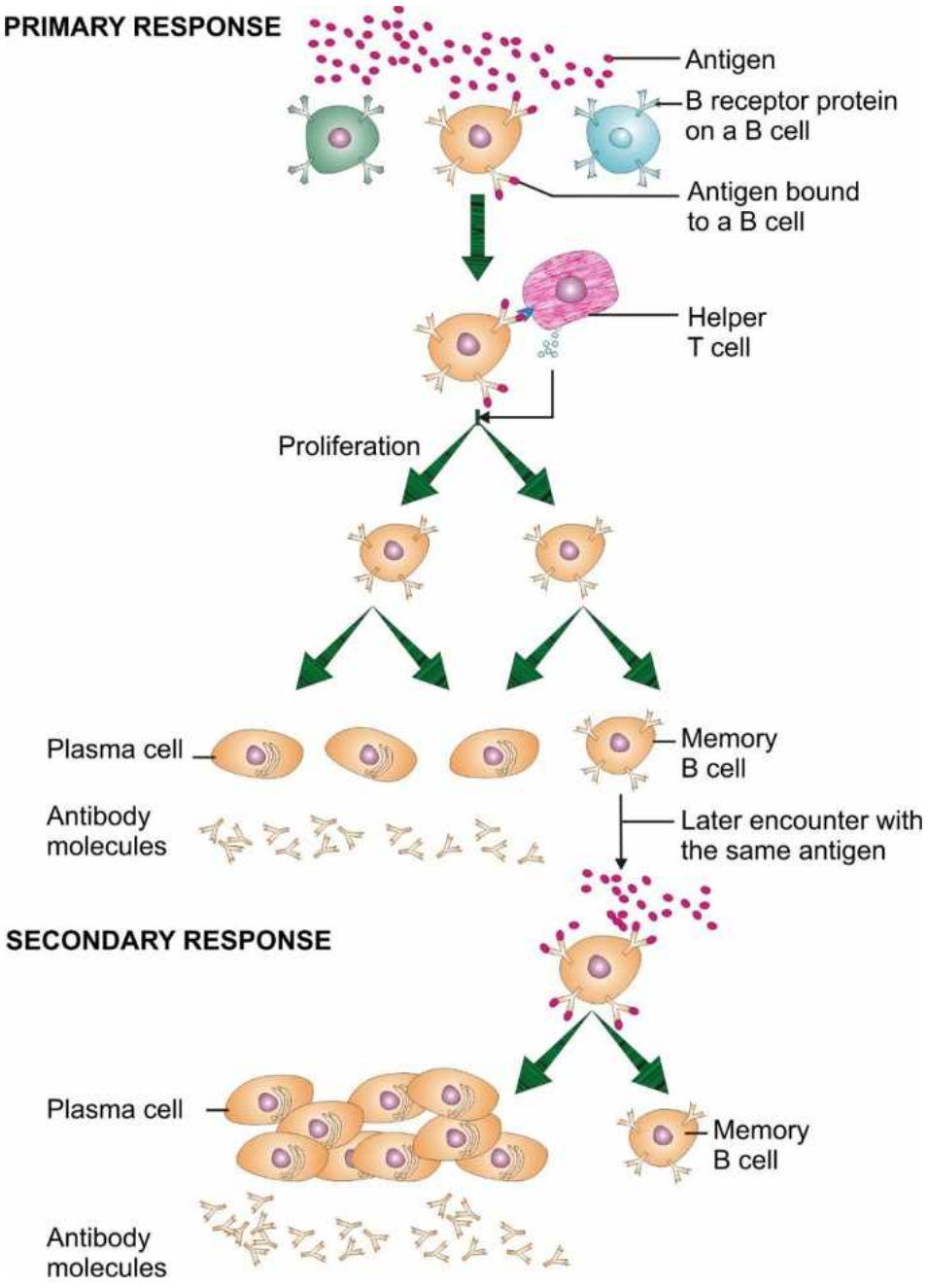


Figure 21.13: Events in antibody-mediated immunity

Structure of Antibody

An antibody is a Y-shaped protein. It is composed of four polypeptide chains; two identical long (heavy) chains and two short (light) chains. The four chains are held together by disulfide bridges. Each chain has a constant region (similar in all the antibodies of the same class) and a variable region (differs among antibodies). The variable regions on one heavy and one light chain of each arm of Y form a specific binding site. Different antibodies have different variable regions. That is why, antibodies are specific i.e., cause the destruction of specific antigen that stimulated their production.

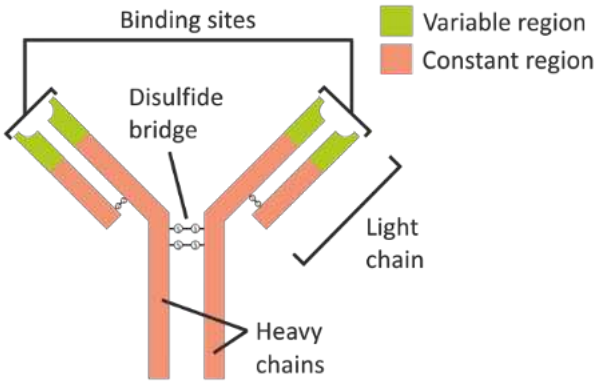


Figure 21.14: Structure of antibody

Monoclonal Antibodies

Only a small portion of antigen molecule fits into an antibody's binding site. This portion is called **determinant**. In some cases, many different antibodies can attach with a typical antigen. In such cases, each antibody fits into a different portion of antigen surface. This type of antibody is called **polyclonal antibody**.

On the other hand, there are antibodies which can attach with only one determinant. These are called **monoclonal antibodies**. In 1984 Cesar Milstein and George Kohler were awarded Nobel Prize for learning how to produce a monoclonal antibody. In their experiment, they mixed plasma cells (that produce antibodies) with cancer cells, called myelomas. They found some new cells growing and dividing in the culture. These cells, called **hybridomas**, used the genes of the plasma cell to make antibodies

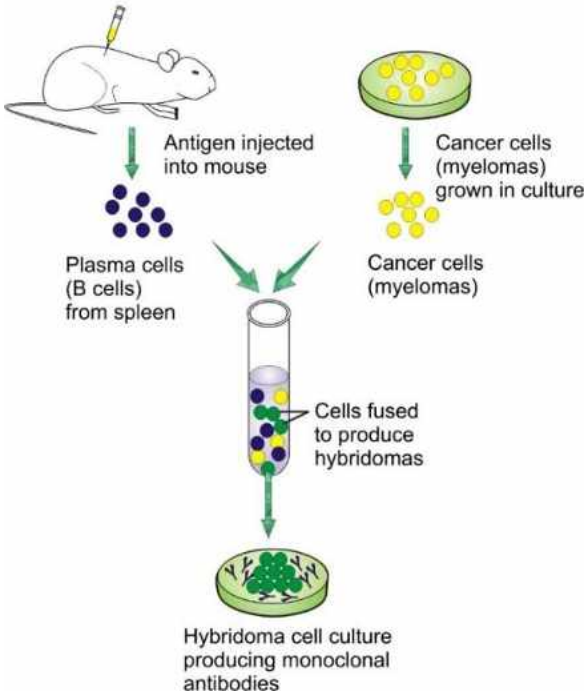


Figure 21.15: Monoclonal antibodies

and used myeloma genes to grow and divide. The hybridoma cells continued to produce the antibody in which the plasma cells had specialized. They obtained rapidly growing cell hybridoma cells which were producing monoclonal antibodies.

Monoclonal antibodies have proved very important, because they can be used to purify specific molecules from complex mixtures. Interferon was purified in this way. In medicine, monoclonal antibodies are used as vehicles for delivering specific therapies. There is a research underway for antigens that occur only on cancer cells, against which radioactive monoclonal antibodies could be targeted to selectively kill cancer cells.

21.3.3- Disorders of Immune System

1- Allergies

Allergies result from the hypersensitivity of immune system to weak antigens that do not cause an immune response in most people. Allergens are the substances that cause allergies e.g., dust, molds, pollen, cat dander, certain foods, and some medicines (such as penicillin).

After exposure to an allergen, some people make a kind of antibody called **E-antibodies**. These antibodies bind to mast cells (usually found in connective tissues surrounding blood vessels). Mast cells release **histamines**, which start the inflammatory response. It causes dilation of blood vessels, leakage of fluid, and other responses changes like sneezing, runny nose, itching etc. In some individuals the histamines can causes life-threatening anaphylactic shock, in which swelling makes breathing difficult. A treatment of common allergies includes antihistamine drugs that block histamines and give temporary relief. Many allergies are also treated by injecting the extracts of the antigens in patients. This treatment is called **desensitization**. This method produces antibodies called **G-antibodies** in the blood. When an allergen enters, it is readily killed by G-antibodies before facing E-antibodies.

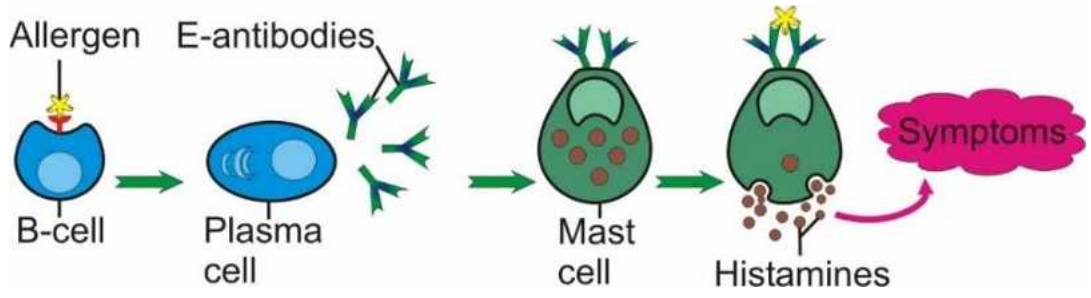


Figure 21.16: Events of allergic response

2- Autoimmune Diseases

Autoimmune diseases result when the immune system is unable to distinguish the body-cells and foreign materials. In this case, the immune system attacks and destroys cells and tissues of the body. The following are some autoimmune diseases.

- **Myasthenia gravis (MG** is a muscle weakness in which patients produce antibodies against acetylcholine receptors on their own skeletal muscle cells. The antibodies bind with the muscle receptors. So, the muscles do not respond to acetylcholine. The nerve impulse cannot communicate the muscle.
- **Systemic Lupus Erythematosis (SLE)** is an autoimmune disease in which the patient forms antibodies against their own connective tissues and major organs.
- **Rheumatoid arthritis:** In this type of arthritis, the patients forms antibodies against the cartilaginous tissue of joints and damage them.
- **Type I diabetes (Juvenile diabetes)** is an autoimmune disease in which antibodies destroy the insulin-producing cells in the pancreas. Most medicines for treating autoimmune diseases suppress immunity in general.

2- Transplant Rejection

Normally the T and B cells of immune system attack and kill only the foreign molecules and cells. It is due to the presence of **self-proteins** on cell surfaces of each person. A particular set of **self-proteins** is like the molecular "fingerprints" recognized by own T and B cells.

Identical twins have a 100% self-proteins match. The best matches are going to occur within a family. The preference order for transplants is identical twin > sibling > parent > unrelated donor. Chances of an unrelated donor matching the recipient are 1 in 100,000-200,000.

When a person receives an organ transplant, there are chances that the self-proteins present on the donated organ do not match the self-proteins of the recipient. As a result, recipient's T cells may attack the cells of the transplanted organ and B cells produce antibodies against its proteins. This problem is called as transplant rejection. To minimize rejection, doctors look for a donor whose self-proteins are matching the recipient's self-proteins, as closely as possible. They also use drugs to suppress the recipient's immune system against the transplanted organs. But these drugs also reduce the ability to fight infections.

There are two approaches to prevent transplant rejection. In one approach, monoclonal antibodies are used to destroy the T cells that attack the transplant. In another approach, stem cells are used to make a new immune system that recognizes the transplant as self.

EXERCISE

SECTION 1: MULTIPLE CHOICE QUESTIONS

1. Which of these are known for stimulating an immune response?
   (a) Pathogens (b) Antigens
   (c) Antibodies (d) Histamines
2. Which of the following is a part of the innate immune system?
   (a) T lymphocytes (b) B lymphocytes
   (c) Skin and mucous membranes (d) Antibodies
3. Which of these is not involved in body's non-specific defence?
   (a) Natural killer cells (b) Inflammatory response
   (c) Antibodies (d) Complement system
4. Skin offers an impenetrable barrier to microbes. It is categorized as;
   (a) Non-specific defence (b) Specific defence
   (c) Acquired immunity (d) Passive Immunity
5. What is the difference between B cells and T cells?
   (a) B cells make antibodies, and T cells destroy infected cells
   (b) B cells and T cells do the same work
   (c) B cells attack germs directly, and T cells make antibodies
   (d) B cells fight germs in body fluids, and T cells work inside cells
6. How do the neutrophils kill microbes?
   (a) By engulfing and digesting them (b) By producing antibodies
   (c) By releasing hormones (d) By carrying oxygen to cells
7. Which group of chemicals is involved in inflammatory response?
   (a) Antibodies (b) Histamines and prostaglandins
   (c) Prostaglandins and pyrogens (d) Antibodies and pyrogens
8. Which type of white blood cell is responsible for producing antibodies?
   (a) Neutrophils (b) B lymphocytes (B cells)
   (c) Macrophages (d) Natural killer cells
9. What types of drug are not prescribed to a recipient of organ-transplantation?
   (a) Drugs that suppress immunity (b) Drugs that strengthen immunity
   (c) Anti-inflammatory drugs (d) Anti-histamine drugs
10. Cell-mediated immunity works best against;
    (a) Viruses infected cells and toxic molecules
    (b) Viruses and toxic molecules
    (c) Virus infected cells and cancer cells
    (d) Toxic molecules and cancer cells

SECTION 2: SHORT QUESTIONS

1. Define the roles of the different types of T cells.
2. Skin is an important component of our defence system. Comment.
3. State how the digestive and respiratory tracts play a role in body's defence.
4. How the natural killer cells play role in body's defence?
5. Define pyrogens and state their effect of hypothalamus.
6. What do you mean by autoimmune diseases?
7. Differentiate between;
   - Antigen and antibody
   - B cells and T cells
   - Macrophages and neutrophils
   - Complement system proteins and interferons
   - Innate and acquired immunity
   - Active and passive immunity
   - Natural active immunity and artificial active immunity
   - Cell-mediated and Antibody-mediated immunity

SECTION 3: LONG QUESTIONS

1. Describe the role of macrophages and neutrophils in killing bacteria.
2. Explain how the proteins of the complement system kill bacteria and how the interferons inhibit the ability of viruses to infect cells.
3. Inflammation is one of the most generalized nonspecific defences. How?
4. How are monocytes, T-cells and B-cells the components of the immune system?
5. Differentiate the two types of acquired immunity (active and passive immunity).
6. Describe the roles T-cells in cell-mediated immunity.
7. Describe the role of B-cells in antibody-mediated immunity.
8. Define allergies. Correlate the symptoms of allergies with the release of histamines.
9. How are T-cells and B-cells related to transplant rejections?

INQUISITIVE QUESTIONS

1. Rationalize the inflammatory response in arthritis as an example of a misdirected immune response.
2. Justify why the physicians prescribe antipyretic drugs, when fever is a nonspecific defence against microbial infections.
3. Justify why physicians prescribe antihistamine therapy to the patients of runny nose or skin rashes.
4. Describe the discovery of monoclonal antibodies and justify how this accomplishment revolutionized many aspects of biological research.

Chapter 22

BIOSTATISTICS

STUDENTS'LEARNING OUTCOMES

After studying this chapter, the students will be able to:

- Define biostatistics and its use.
- Define mean, median, mode, standard deviation, range and percentile.
- Calculate mean, median, mode, standard deviation, range and percentile from a given set of data.
- Sketch a bar chart from a given set of data.
- Sketch error bars based off of range or standard deviation for a given set of data on a bar chart.
- Evaluate the appropriate type of figure or chart for a given set of data and/or experiment (bar chart, pie chart, x-axis data figure etc.).
- Make the appropriate chart with proper title, labelled axis, legend, axis units.
- Design an appropriate experiment with a control group and dependent, independent and control variables.

22.1 BIOSTATISTICS

Biostatistics is a scientific discipline that applies statistical methods and techniques specifically to biological, medical, and health-related data. It involves the collection, presentation, analysis, and interpretation of data to draw meaningful conclusions. Biostatistical Analysis allows researchers and health professionals to make informed decisions about populations using sample data, rather than testing entire populations which may be impractical or costly. For example, by studying a representative subset of workers in a population, biostatistics helps estimate the prevalence of a health condition among all workers.

22.1.1 Components of Biostatistics

The key components of biostatistics comprehensively encompass several critical areas that collectively enable the proper collection, analysis, interpretation, and application of biological or health-related data. These components include:

1- Study Design and Experimental Planning

This involves formulating the research question, determining objectives, and structuring experiments or observational studies to test hypothesis. It includes selecting the population, defining variables, randomization, controlling confounding factors, and calculating adequate sample size. Proper study design ensures validity, reliability, and unbiased results.

2- Data Collection

Collecting accurate and relevant data is fundamental. This can be done through various methods:

Primary Data Collection: Direct data from experiments, surveys, interviews, observations, and clinical trials.

Secondary Data Collection: Using existing data from published research, medical records, government reports, and databases.

3- Data Management and Processing

Organizing the collected data systematically and ensuring accuracy before analysis. It includes data cleaning, coding, and storage, maintaining confidentiality and integrity.

4- Data Analysis

Application of statistical techniques to summarize, describe, and draw inferences from data. **Descriptive statistics** describe characteristics of the data (mean, median, mode, standard deviation etc.) **Inferential statistics**, including hypothesis testing, regression analysis, and modelling, allow conclusions about populations from sample data.

5- Interpretation of Results

Translating statistical findings into meaningful biological or health conclusions. Consideration of context, limitations, and assumptions of the analysis is important.

6- Presentation and Communication

Effectively communicating results using visualizations (charts, graphs) and clear language tailored to scientific, medical, and public audiences. Transparency in reporting methods and findings is essential for reproducibility and trust.

22.1.2 Applications of Biostatistics

1. Clinical trials and medical research

Biostatistics plays a crucial role in clinical trials and medical research by providing the framework for designing studies and analyzing data to ensure valid and reliable outcomes. Through statistical methods, biostatisticians evaluate the safety and effectiveness of new drugs, treatments, and medical devices. This process enables healthcare professionals to make evidence-based decisions that improve patient care and advance medical knowledge.

2. Epidemiology

In epidemiology, biostatistics is used to investigate the distribution and determinants of diseases within populations. It helps public health experts understand how diseases spread, identify risk factors, and evaluate the impact of preventive measures. By modelling disease trends and assessing intervention

outcomes, biostatistics supports efforts to control epidemics and improve community health.

3. Genetics and genomics

The field of genetics and genomics relies heavily on biostatistics to analyze complex genetic data. This includes uncovering relationships between genes and diseases, understanding inheritance patterns, and exploring genetic variations. Biostatistical methods are essential in personalized medicine, where treatments are tailored to an individual's genetic profile to enhance effectiveness and reduce adverse effects.

4. Public health policy

Biostatistics is instrumental in shaping public health policy by analyzing population health data and evaluating health programs. It identifies vulnerable groups and health disparities, providing policymakers with evidence to design and implement interventions that promote equity and improve overall health outcomes on a large scale.

5. Environmental health

Environmental health benefits from biostatistics through the assessment of how environmental factors like pollution and toxins affect human health. Statistical analysis helps identify harmful exposures and guides regulations and preventive strategies to protect communities from environmental hazards.

6. Bioinformatics

In bioinformatics, biostatistics supports the analysis of vast biological datasets from genomics and proteomics. By developing algorithms and models, biostatisticians enable the interpretation of complex data essential for drug discovery, disease diagnosis, and the development of personalized therapies.

7. Healthcare quality control

Healthcare quality control also relies on biostatistics to monitor and improve healthcare services. By applying statistical tools, healthcare providers can evaluate diagnostic test accuracies, track patient outcomes, and ensure consistency in medical procedures, ultimately enhancing the quality of patient care.

8. Health programs and population interventions

Biostatistics evaluates the effectiveness of health programs and population-based interventions. It helps public health officials understand the impact of immunization campaigns, health education efforts, and training programs for healthcare workers. This evaluation is vital for optimizing resource allocation and maximizing the benefits of public health initiatives.

9. Controlling epidemics

During epidemics, biostatistics is critical in tracking the spread of disease, estimating mortality rates, and identifying at-risk populations. This real-time statistical analysis informs decision-making and helps implement targeted strategies to contain outbreaks and mitigate their effects.

10. Identifying barriers to care

Biostatistics also uncovers barriers to healthcare access by analyzing survey data and health service usage patterns. This insight helps healthcare providers and policymakers develop strategies to improve access and reduce health disparities, making healthcare systems more efficient and equitable.

11. Health risk assessment and demography

Finally, biostatistics plays an essential role in health risk assessment and demography by analyzing trends in births, deaths, disease prevalence, and other vital statistics. These analysis provide crucial information for government agencies and health organizations to plan services, allocate resources, and address public health challenges effectively. Together, these multifaceted uses illustrate the indispensable nature of biostatistics in improving health outcomes worldwide.

22.2 KEY STATISTICAL MEASURES AND THEIR CALCULATION

In the field of statistics, understanding and calculating key measures is essential for summarizing and interpreting data. These measures provide valuable insights into the central tendency, variability, and distribution of data, helping to identify patterns and make informed decisions. The mean represents the average value, the median indicates the middle point, and the mode highlights the most frequent value of dataset. Standard deviation and range measure the spread and dispersion of data, while percentiles break the data into specific intervals for a deeper analysis.

22.2.1 Mean

The mean, also known as the average, is a measure of central tendency that gives an idea of the central value in a dataset. From a biological perspective, it can be used to summarize large datasets, providing a central value that represents typical characteristics, such as average size or growth rate.

Formula

For ungrouped data

$$\text{Mean} \quad \frac{x}{n}$$

Where,

x = sum of all the values.

n = number of values in the dataset.

For grouped data

$$\text{Mean} = \frac{\sum_{i=1}^{n} f_i x_i}{\sum_{i=1}^{n} f_i}$$

Where:

x_i = midpoint of the ith class interval.

f_i = frequency of the ith class.

f_i = sum of all the frequencies.

Steps to calculate the Mean (for ungrouped data)

A biologist measured the average weight of a specific species of bird in a park. He recorded the weights (g) of five birds from the same species. The bird's weights are 400g, 420g, 470g, 390g and 460g.

Step 1: Add all the weights together (X)

$$X = 400+420+470+390+460 = 2140 \text{ g}$$

Step 2: Count the total number of birds (n)

$$n = 5 \text{ birds}$$

Step 3: Divide the sum of bird's weights by the number of birds

$$\text{Mean} = \frac{X}{n} = \frac{2140}{5} = 428g$$

The average weight of a specific species of birds is 428g.

Steps to calculate the Mean (for grouped data)

A biologist studied the growth of a certain plant species under different soil conditions. The plant heights (cm) are grouped into class intervals based on measurements from different plots of land with varying soil types.

Step 1: Organize the data set into class intervals.

Step 2: Calculate the midpoint of each class interval.

Step 3: Multiply each midpoint by its corresponding frequency.

Step 4: Sum the products of midpoints and frequencies.

Step 5: Divide the result of the step 4 by the total frequency.

Table: Mean plant height (cm) calculation using grouped data across soil types.

Soil Type	Class Interval (Height in cm)	Frequency (f_i)	Midpoint (x_i = lower limit + upper limit / 2)	f_i, x_i
Sandy Soil	0 – 10	5	5	25
Loamy Soil	10 – 20	8	15	120
Clayey Soil	20 – 30	7	25	175
Peaty Soil	30 – 40	4	35	140
Median		$f_i = 24$		$f_i x_i = 4601$

$$\text{Mean} = \frac{\sum f_i x_i}{\sum f_i}$$

$$= \frac{460}{24} = 19.16667 \approx 19.17(cm)$$

The average growth of a certain plant species under different soil conditions is 19.17cm.

22.2.2 Median

The median is a measure of central tendency that represents the middle value in a dataset when the data is arranged in order. If the number of values in data set is odd, the median is the middle values. If the number of values in data set is even, the median is the average of the two middle numbers. From a biological perspective, it provides a better representation of the central tendency in cases like population size, disease prevalence, or body measurements, where data might not be normally distributed.

Formula

There is no single formula to calculate the median but there are steps to calculate it.

Steps to calculate the Median

Step 1: Arrange the dataset in ascending or descending order.

Step 2: Count the total number of values in the dataset.

Step 3: If the number of values in the dataset is odd, the median is the value at the position $\left(\frac{n+1}{2}\right)^{th}$.

Step 4: If the number of values in the dataset is even, the median is the average of values at the positions $\left(\frac{n}{2}\right)^{th}$ and $\left(\frac{n+1}{2}\right)^{th}$.

Example 1 (odd number of values)

A biologist studied the average height (cm) of five plants in a garden. The heights of the plants are 50cm, 65cm, 60cm, 75cm, and 80cm.
Step 1: 50cm, 60cm, 65cm, 75cm and 80cm.
Step 2: n =5 plants
Step 3: n =5 is odd, the median is the value at the position $\left(\frac{5+1}{2}\right)=\left(\frac{6}{2}\right)=3$.

Example 1 (even number of values)

A biologist studied the average weight (kg) of six different animals in a zoo. The weights of the animals are 300kg, 400kg, 330kg, 315kg, 450kg and 500kg.
Step 1: 300kg, 315kg, 330kg, 400kg, 450kg and 500kg.
Step 2: n =6 animals
Step 3: n = 6 is even, the median is the average of value at the position $\left(\frac{6}{2}\right)=3$ and $\left(\frac{6+2}{2}\right)=\left(\frac{8}{2}\right)=4$.

Median= 330 + 400/2=365kg

22.2.3- Mode

The mode is the value or values that appear most frequently in a dataset. From a biological perspective, it can be used to understand the most common characteristics or traits in a biological sample, such as the most frequent number of flowers on a plant, the most common size of seeds in a population, or the most frequently occurring weight in a sample of animals.

Formula

There is no specific formula to calculate mode in **ungrouped data**. The mode is simply the value or values that appear more frequently in a data set.
For **grouped data** the mode can be calculated by using the given formula:

$$\text{Mode} = l + \frac{(fm - f_1)}{(fm - f_1) + (fm - f_2)} \times h$$

Where:
l = lower boundary of the modal class.
fm = frequency of the model class.
F_1 = frequency of the class before the modal class.
f_2 = frequency of the class after the modal class.
h = class width (the difference between upper and lower limits of any class interval).

Steps to calculate the Mode for ungrouped data with example

A biologist studied the height (cm) of ten plants in a garden. The heights of plants are 50cm, 60 cm, 50 cm, 55 cm, 50 cm, 65 cm, 60 cm, 70 cm, 55 cm and 50 cm.

Step 1: Arrange the data in ascending order.

50 cm, 50 cm, 50 cm, 50 cm, 55 cm, 55 cm, 60 cm, 60 cm, 65 cm, 70 cm.

Step 2: Count the frequency of each value.

50 cm appears 4 times
55 cm appears 2 times
60 cm appears 2 times
65 cm appears 1 time
70 cm appears 1 time

Step 3: Identify the mode.

The number 50 appears the most frequently i.e. 4 times. Therefore, the mode for this dataset is 50 cm.

Steps to calculate the Mode for grouped data with example

Consider we are studying the weight (kg) distribution of animals in a zoo.

Class	Class interval Weight (kg)	Frequency
I	10-20	5
II	20-30	12
III	30-40	18
IV	40-50	10
V	50-60	3

Step 1: Identify the modal class.

The class interval (30-40) Class III is the modal class with the highest frequency (18).

Step 2: Put the values in the formula to calculate mode for grouped data.

l =30 (lower boundary of the modal class)
fm =18 (frequency of the modal class)
f_1 =12 (frequency of the class before the modal class)
f_2 =10 (frequency of the class after the modal class)
h =10 (class width)

$$\text{Mode} = l + \frac{(fm - f_1)}{(fm - f_1) + (fm - f_2)} \times h$$

$$= 30 + \frac{(18 - 12)}{(18 - 12) + (18 - 10)} \times 10$$

$$= 30 + \frac{6}{6 + 8} \times 10 = 30 + \frac{6}{14} \times 10$$

$$= 34.29\, kg$$

The mode of the givens dataset is 34.29 kg.

22.2.4- Standard deviation

A standard deviation is a measure of how dispersed the data is in relation to the mean. Low, or small, standard deviation indicates data are clustered tightly around the mean, and high, or large, standard deviation indicates data are more spread out. A standard deviation close to zero indicates that data points are very close to the mean, whereas a larger standard deviation indicates data points are spread further away from the mean.

a) **The curve is more spread out and has a high standard deviation**	b) **The curve is more clustered around the mean and has a lower standard deviation**

In biology, standard deviation is commonly used to assess the variability in measurements, such as the growth rates of plants, the concentration of enzymes in a sample, or any other biological data.

Formula

$$\sigma = \sqrt{\frac{\sum\left(x_i - \mu\right)^2}{N}}$$

Where:

σ = standard deviation

x_i = each individual data point in the set

μ = mean

N = total number of data points.

Steps to calculate Standard Deviation with example

A biologist studied the heights of 9 different plants of the same species. The heights of the plants are 56cm, 65cm, 74cm, 75cm, 76cm, 77cm, 80cm, 81cm and 91cm.

Step1: Find the mean.

Step2: Subtract the mean from each data point to find the deviation of each point.

Step3: Square the result of each deviation which eliminates negative values and emphasizes larger deviations.

Step4: Find the average of the squared deviations.

Step5: Take the square root of the result from step 4. This is the standard deviation.

Table: Calculation of standard deviation for plant heights

Height in (cm) x_i	Mean μ	Subtract mean from each data point $(x_i - \mu)$	Square the result of each deviation $(x_i - \mu)^2$	Sum of squared deviations $\sum(x_i - \mu)^2$	Variance (Average of the squared deviations) $\sum(x_i - \mu)^2/N$	Standard deviation $\sigma = \sqrt{\frac{\sum(x_i - \mu)^2}{N}}$
56	75	56 – 75 = -19	361	784	87.11	9.33
65		65 – 75 = -10	100			
74		74 – 75 = -1	1			
75		75 – 75 = 0	0			
76		76 – 75 = 1	1			
77		77 – 75 = 2	4			
80		80 – 75 = 5	25			
81		81 – 75 = 6	36			
91		91 – 75 = 16	256			

22.2.5 Range

The range is a measure of the spread or dispersion of a set of data. It is the simplest form of variability and is used to understand the extent of variation in the dataset. The range represents the difference between the maximum and minimum values in a dataset. A larger range suggests a greater spread of data, meaning the data points are more dispersed from each other while a smaller range indicates that the data points are closely clustered around a central value.

Formula

$$\text{Range} = X_{\max} - X_{\min}$$

Where:

X_{max} = maximum value in the data set.

X_{min} = minimum value in the data set.

Steps to calculate the Range with example

Consider a dataset representing the ages of a group of people: 25, 20, 22, 30, 42, 45, 50.

Step 1: Arrange the data in ascending order.

20, 22, 25, 30, 42, 45, 50

Step 2: Identify the greatest value in the dataset.

50

Step 3: Identify the smallest value in the dataset.

20

Step 4: Subtract the maximum value from the minimum value to calculate the range.

$$\text{Range} = X_{\max} - X_{\min}$$

$$\text{Range} = 50\text{-}20 = 30$$

22.2.6 Percentile

A percentile is a measure that indicates the value in a dataset below which a given percentage of observations in a group of data falls. For instance, the 25th percentile, also known as the first quartile (Q1), is the value below which 25% of the data points lie. Similarly, the 50th percentile, or the median, is the value below which 50% of the data points lie, and the 75th percentile, or the third quartile (Q3), is the value below which 75% of the data points lie. Percentiles are particularly useful in biostatistics as they help in understanding the distribution of biological data which is skewed or normally distributed.

Formula

$$R = R = \frac{P}{100} \quad (n + 1)$$

Where

R = rank

P = desired percentile

n = number of observations in the data set

If R is an integer, the value at the Rth position in the ordered dataset is the percentile. If R is not an integer (e.g., 4.4), interpolation between the two nearest ranks is used to estimate the percentile value. For example, if it lies between 4th and 5th data points, percentile value=

Value at 4th + 0.4 X (value at 5th - value at 4th)

Steps to calculate percentile with example

Suppose we have the following dataset representing the heights (cm) of 10 individuals: 160, 162, 168, 165, 170, 175, 172, 178, 180, 182.

To calculate the 40th percentile:

Step 1: Arrange the data in ascending order.

160, 162, 165, 168, 170, 172, 175, 178, 180, 182.

Step 2: Determine the total number of observations.

n = 10

Step 3: Calculate the rank.

$R = P/100 \times (n+1)$

$R = 40/100 \times (10+1) = 4.4$

Step 4: Locate the rank in the dataset and interpolate if necessary.

The rank is 4.4, which is not an integer. This means that the 40th percentile lies between 4th (168) and 5th (170) data points.

Interpolate between the 4th and 5th values:

Percentile value = value at 4th + 0.4 X (value at 5th - value at 4th)

Percentile value = 168 + 0.4 X (170-168) = 168 + 0.4 X 2 = 168 + 0.8 = 168.8

Step 5: Interpret the result.

The 40th percentile value is 168.8. This means that 40% of the observations fall below or equal to 168.8 in this data set.

22.3 CHART

22.3.1 Bar Chart

A bar chart (or bar graph) is a graphical representation used to display categorical data with rectangular bars, where the length or height of each bar is proportional to the value it represents. Bar charts are used to compare quantities across distinct categories or groups. In biology, bar charts are valuable for showing data like the counts of different species, experimental groups, or measurement results from biological samples. The key characteristics of a bar chart are:

i) Categories are represented by the x-axis (horizontal axis).

ii) Values or frequencies are represented by the y-axis (vertical axis).

iii) Bars can be oriented vertically or horizontally.

Steps to create a bar chart with example

A team of research students conducted an insect diversity study in a local forest and counted the number of individuals for five common insect species over a month.

Table: Relative abundance of the insect species

Insect species	Number of individuals counted	Percentage (%)
Ladybug	45	16
Ant	120	41
Butterfly	30	10
Bee	75	26
Dragonfly	20	7

Step 1: Purpose and data.

Purpose: To visually compare the abundance of five different insect species.

Categorical variable: Insect species (ladybug, ant, butterfly, bee, dragonfly).

Numerical variable: Number of individuals counted.

Step 2: Determine the axis.

X-axis (Categorical): Insect species
Y-axis (Numerical): Number of individuals counted.
Step 3: Scale the numerical axis.
Greatest value is 120 (Ants). Scale the y-axis from 0 to 130 or 140. An increment of 10 or 20 is used for good readability. Let's choose 0 to 140 with increment of 20.
Step 4: Draw the bars.
Draw a bar for ladybug up to 45 on the y-axis.
Draw a bar for ant up to 120 on the y-axis.
Draw a bar for butterfly up to 30 on the y-axis.
Draw a bar for bee up to 75 on the y-axis.
Draw a bar for dragonfly up to 20 on the y-axis.
Ensure bars are of equal width and evenly spaced.
Step 5: Add labels and title.
Chart title: Insect species abundance in local forest
X-axis label: Insect species
Y-axis label: Number of individuals

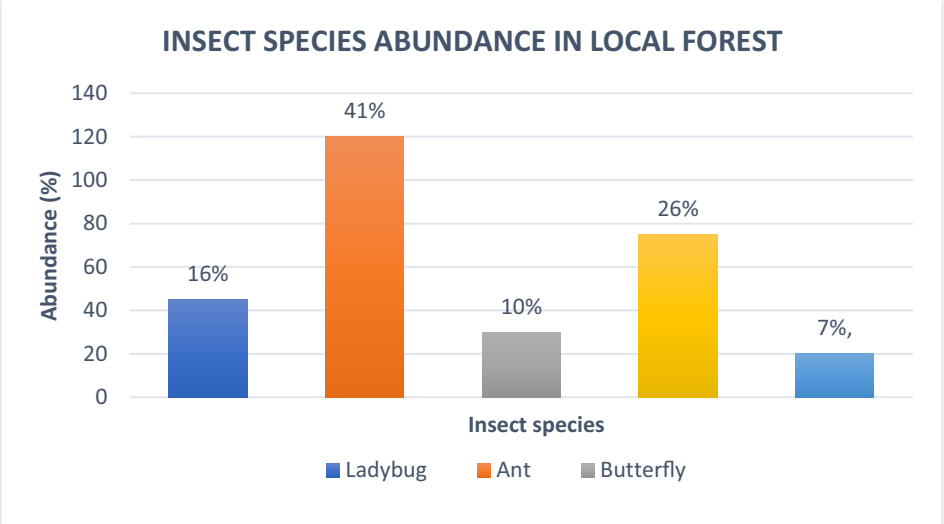


Fig. Bar chart showing relative abundance of insect species in a local forest

A **legend** is a key or guide on a chart, graph, or map that explains the meaning of symbols, colours, patterns, or labels used in the visual. It helps the viewer understand what different elements represent, making the data or information easier to interpret and analyse. For example, in a bar chart with different colours for each category, the legend identifies what each colour corresponds to.

22.3.2 Pie Chart

A pie chart is a circular statistical graph divided into sectors (slices), each representing a proportion of the whole dataset. The entire circle corresponds to 100% (or 360°), and each sector's angle is proportional to its category's share in the dataset. Pie charts are ideal for visualizing categorical data as portions of a whole, making it intuitive to compare different group's contributions in biology, such as types of cells, species distributions, or survey responses.

Steps to create a pie chart with example

A biology class surveys 100 students to find their blood types. The results are as follows:

Table: Surevy results of student blood types	
Blood type	**Number of students (frequency)**
A	30
B	20
AB	10
O	40

Step 1: Organize data

Arrange the categorical data in a table, noting the frequency or total for each category as shown in table.....

Step 2: Add up all frequency values to get the total sum

Total number of students= 30+20+10+40 = 100

Sum of frequency	Blood Type	No. of Student	Sector angle = Sum of values
30	A	30	$\frac{30}{100} \times 360° = 108°$
20+30=50	B	20	$\frac{20}{100} \times 360° = 72°$
10+50=60	AB	10	$\frac{10}{100} \times 360° = 36°$
40+60=100	O	40	$\frac{40}{100} \times 360° = 144°$
	Total	n =100	

Step 3: Calculate the sector angles (Sector angle = Category value/ total value X 100)

A= 30/100 X 360° = 108° %

B= 20/100 X 360° = 72° %

AB= 10/100 X 360° = 36° %

O= 40/100 X 360° = 144° %

Step 4: Draw the pie chart

Draw a circle by using a protractor, draw each sector angle in sequence (clock wise or counter clockwise) using the calculated angles in step 3. Start with a reference line (radius). Use different colours/shades to differentiate each category.

Step 5: Label and title

Clearly label each sector and provide a legend if needed. Add a descriptive title to the chart.

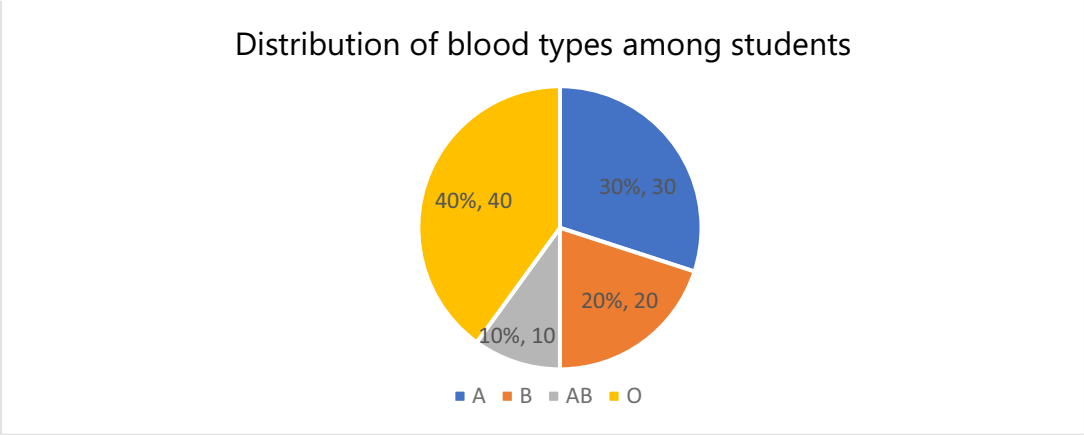


Fig. Pie chart showing distribution of blood types among students

X-axis data figure

The x-axis refers to the horizontal line on a Cartesian coordinate system or data figure such as a graph, chart, or plot. It is most commonly used to represent the independent variable. The x-axis provides the baseline against which other variables (typically shown on the y-axis) are measured. It is always drawn horizontally, running left to right, and often starts at the origin (0, 0), extending in the positive direction, though it can also include negative values when needed. In biological studies, the *x*-axis is often used to represent variables like time intervals, different species, or experimental conditions. It provides a **baseline for comparison** and helps visualize **trends or relationships** between variables in biological data.

Steps to create an x-axis data figure with example

Suppose we are measuring the activity of enzyme. The activity is measured at 5°C, 10°C, 20°C, 30°C and 40°C.

Step 1: Identify variables

Independent variable (x-axis): Temperature, measured in degrees Celsius (°C).

Dependent variable (y-axis): Rate of enzyme activity

Step 2: Determine scale and range

Decide the minimum and maximum values, and the intervals or categories for the x-axis. Temperatures at which enzyme activity is measured i.e. 5°C, 10°C, 20°C, 30°C, and 40°C.

Step 3: Label the axis

X-axis is labeled as Temperature (°C) and Y-axis is labeled as Enzyme Activity (µmol/min).

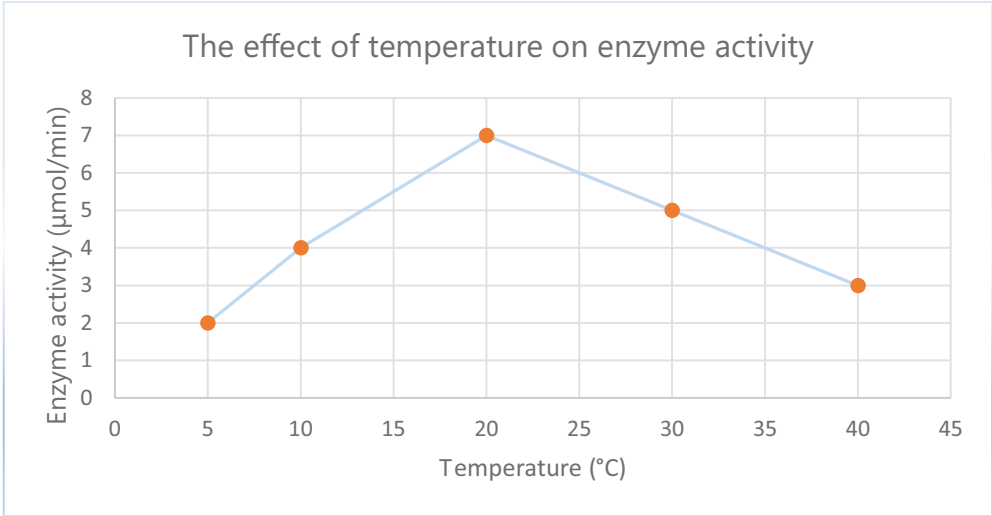
Step 4: Plot the data points

For each measurement, find its position on the x-axis and plot it against the corresponding y-axis value (the dependent variable). We measured these pairs: (5°C, 2 µmol/min), (10°C, 4 µmol/min), (20°C, 7 µmol/min), (30°C, 5 µmol/min), (40°C, 3 µmol/min).

Step 5: Adjust formatting

Make sure tick marks are evenly placed for this data. Temperature values are evenly spaced along the x-axis since the interval between points is similar. Connect the data points with a line to show the trend.

Fig. X-axis data figure showing the effect of temperature on enzyme activity



Error Bars

Error bars are graphical elements placed on bar charts (and other types of plots) to visually represent the variability or uncertainty in data. They help visualize how much the individual data points typically deviate from the central tendency (e.g., mean) represented by the bar.

Types of error bars

The two most common ways to sketch errors bars for a bar chart are:

Range error bars

Range error bars use the smallest (minimum) and greatest (maximum) data points in a set for each bar. The bar extends from the minimum to the maximum value, illustrating the full spread of the data.

Standard deviation (SD) error bars

Standard deviation error bars show the average distance of each data point from the mean. On a bar chart, each bar represents a group mean. **Standard deviation error bars** extend **above and below the mean** value of a dataset by one or more standard deviations (±SD).

Steps to sketch error bars on a bar chart

The researchers are measuring the average plant height (cm) in three different light conditions. They grow bean plants in three groups:

Group A: Low light
Group B: Medium light
Group C: High light

After four weeks they measure the heights of the plants.

	Group A	Group B	Group C
Plant 1	10	14	18
Plant 2	11	16	20
Plant 3	9	15	19

Step 1: Calculate the mean of each group
Group A Mean of = (10 + 11 + 9)/3 = 10 cm
Group B Mean of= (14 + 16 + 15)/3 = 15 cm
Group C Mean of= (18 + 20 + 19)/3 = 19 cm
The bars on the bar chart are drawn at heights 10, 15, and 19 for Groups A, B, and C, respectively.

Step 2: Calculate the spread

A) For SD error bars:
Group A: SD ≈ 1 (Data: 10, 11, 9)
Group B: SD ≈ 1 (Data: 14, 16, 15)
Group C: SD ≈ 1 (Data: 18, 20, 19)

B) For range error bars:
Group A: min = 9, max = 11
Group B: min = 14, max = 16
Group C: min = 18, max = 20

Step 3: Draw the error bars

SD error bars: Start at the mean (top of each bar), extend up and down by 1 cm (Mean ±1 SD).
Group A: from 9 (10-1) to 11 (10+1)
Group B: from 14 (15-1) to 16 (15+1)
Group C: from 18 (19-1) to 20 (19+1)

Range error bars: Extend from the minimum to maximum value for each group. Add a horizontal cap at each end of the vertical error bar for clarity.

Table: Statistical summary of plant measurements

Group	Plant 1	Plant 2	Plant 3	Mean	Range (max-min)	Range error (±)= Range/2	Standard deviation error(±SD)
A	10	11	9	10	2	1	1
B	14	16	15	15	2	1	1
C	18	20	19	19	2	1	1

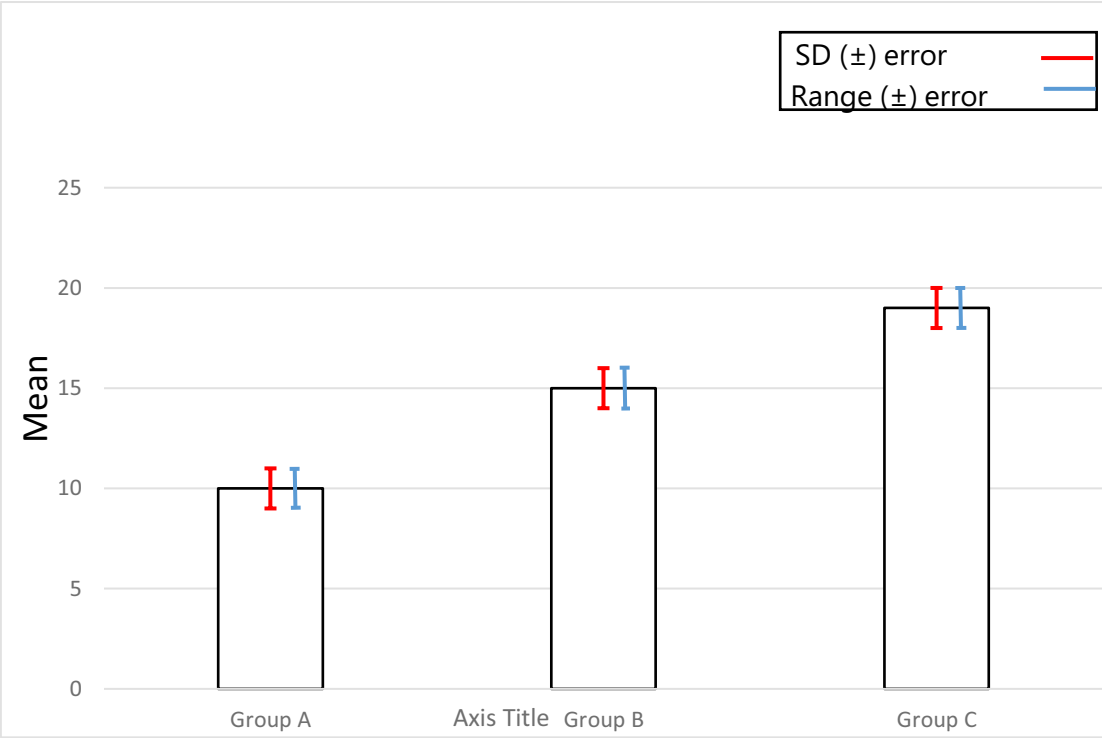


Fig. Graph showing average plant height in different light conditions with error bars

22.3 Experimental Design

When scientists want to test an idea, they plan an experiment carefully. This planning is called **experimental design**. They decide subjects or participants and control the conditions so results are fair. Then, they change one thing (the independent variable) and see how it affects another thing (the dependent variable). This help them find out if their hypothesis is right or wrong by looking at how different variables are connected.

To do this effectively, scientists identify different types of variables. The **independent variable** is the factor that the experimenter changes or manipulates to observe its effect. The **dependent variable** is what is measured or observed; it changes in response to the independent variable. **Control variables** (or constants) are all the other factors that must be kept the same throughout the experiment to ensure a fair test.

A **control group** is a group in the experiment that does not receive the treatment or change in the independent variable. It serves as a baseline to compare the results of the experimental group, helping to show whether the independent variable actually causes any effect. An **experimental group** is a group that is exposed to the specific treatment, condition, or environmental change being tested in an experiment. Their responses are measured and compared to a control group to assess the effect of that treatment on biological processes or characteristics.

Steps for experimental design:

1. State the question of your experiment first. This clearly defines what you want to find out.
2. Formulate a hypothesis based on your research e.g., what you think will happen, based on existing knowledge.
3. Clearly identify variables to understand what you will change (independent variable), measure (dependent variable) and keep constant (control variable).
4. Set up groups e.g., control group (does not receive the treatment or change in independent variable) and experimental group (receives the treatment or change).
5. Plan the procedure and outline detailed steps on how experiments will be conducted e.g., how subjects are selected and assigned to groups, how each group treated, the duration and frequency of measurement and how data will be collected.
6. Use appropriate methods (such as averaging, graphing or statistical tests) to compare outcomes between groups and assess whether there is a significant effect
7. Draw conclusions in relation to the original hypothesis and research question. Accept or reject hypothesis on the basis of results.

Experiment: Effect of high-fat diet on weight gain in albino mice

1.	**Question:**	Does a high-fat diet cause greater weight gain in mice compared to normal diet?
2.	**Hypothesis:**	Mice given a high-fat diet will gain more weight than those on a normal diet.
3.	**Identify variables:**	**Independent variable:** Type of diet **Dependent variable:** Weight gain **Control variables:** Mouse age, strain, sex, cage, environment (light, temperature) and feeding schedule etc
4.	**Set up groups:**	**Control group:** Mice on normal diet **Experimental group:** Mice on a high-fat diet
5.	**Procedure outline:**	1. Select albino mice that are matched by age, strain, and sex. 2. Randomly assign albino mice to control and experimental groups. 3. House all albino mice under identical conditions (same cages, light, temperature, humidity). 4. Administer the assigned diet to each group for a set period (e.g., 8 weeks). 5. Measure and record each albino mice weight weekly.
6.	**Analyse results:**	Use statistical tests or graphs to compare the average weight gain between groups at the end of the experiment.
7.	**Draw conclusions:**	The experiment will determine whether a high-fat diet leads to greater weight gain in mice compared to a normal diet. If the high-fat diet group shows a statistically significant increase in average weight gain, the hypothesis will be supported, suggesting that diet type influences weight gain. However, if there is no significant difference between groups, or if the normal diet group gains more weight, the hypothesis will be rejected, indicating that factors other than diet type may play a greater role in influencing weight gain under the given experimental conditions.

EXERCISE

SECTION 1: MULTIPLE CHOICE QUESTIONS

1. Which of the following is a primary data collection method?
   (a) Government reports (b) Published articles
   (c) Experiment and surveys (d) Online databases
2. What does mode represent in a dataset?
   (a) The average of values (b) The value occurring most frequently
   (c) The difference between two values (d) None of these
3. Which statistical measure is also called the 50th percentile?
   (a) Mean (b) Median
   (c) Mode (d) Range
4. If the maximum value in a dataset is 50 and the minimum value is 20, what is the range?
   (a) 30 (b) 25
   (c) 20 (d) 115
5. What does it mean if the 40th percentile value is 148cm?
   (a) 40% of values are above 148cm
   (b) 40% of values are below or equal to 148cm
   (c) All the values are equal to 148cm
   (d) 60% of values are above 148cm
6. In a pie chart, what does the full circle represent?
   (a) 180° (b) 280° (c) 380° (d) 360°
7. What do error bars represents in a chart?
   (a) Average (b) Variability or uncertainty in data
   (c) Both of these (d) None of these
8. In an experiment, what does the dependent variable represent?
   (a) The factor kept constant (b) The factor being changed
   (c) The factor ignored (d) All of these
9. What is the main purpose of including a control group in an experiment?
   (a) To test the dependent variable (b) To provide a baseline for comparison
   (c) To collect random data (d) All of these
10. Which group in an experiment receives the treatment?
   (a) Control group (b) Dependent group
   (c) Experimental group (d) Independent group

SECTION 2: SHORT QUESTIONS

1. Explain why biostatistics is important in medical research?
2. Describe the difference between descriptive and inferential statistics?

3. What is the relationship between percentiles and quartiles?
4. How are independent and dependent variables related in experimental design?
5. The height of six children are 110, 115, 118, 120, 122, and 125. What is the median height of the children?
6. What does a legend in a chart present?
7. What are error bars?

SECTION 3: LONG QUESTIONS

1. Explain the concepts of mean, median, and mode as measures of central tendency. Illustrate with examples when each measure would be most appropriate.
2. Describe standard deviation, range, and percentiles as measures of variability. Compare their advantages and limitations in interpreting biological data?
3. What are error bars? Explain their types (range error bars, standard deviation error bars) and significance in experimental biology. Illustrate your answer with a hypothetical dataset?
4. Compare and construct bar charts and pie charts. Which situations favour one over the other? Support your answer with real-world biological or health data examples.
5. Discuss different types of data presentation tools in biostatistics (bar chart, pie chart and error bars). For each, explain an appropriate biological example where it would be the best choice.

INQUISITIVE QUESTIONS

1. A pharmaceutical company claims their drug improves recovery time from flu. Propose an experimental design using biostatistics principles, and explain how you would ensure validity and reliability of results?
2. Evaluate the usefulness of range versus standard deviation in representing variability. Which one would you trust more in medical research and why?
3. During an epidemic, mortality rate estimates vary depending on whether the mean, median, or percentile is used. Discuss how policymakers might misinterpret such statistics and how biostatistics can prevent this.

Chapter 23

PHARMACOLOGY

STUDENTS'LEARNING OUTCOMES

After studying this chapter, the students will be able to:

- Explain the drug discovery and development process.
- Define 4 classes of antibiotics (penecillins, Tetracyclins, Fluriquinolones and Sulfonamides) and describe their mode of action
- Define antivirals and antiretrovirals
- Describe advantages of monoclonal antibodies as compared to other drug classes.

Pharmacology is the branch of biology that deals with the study of drugs and their effects on living organisms. It includes the study of the sources, properties, composition, therapeutic uses, and effects of drugs on the body. Drugs are chemical substances used to diagnose, prevent, or treat diseases.

Drugs may be obtained from natural sources such as plants, animals, and microorganisms, or they may be synthesized in laboratories. Medicines prepared from plants are commonly used in traditional systems of treatment and are known as herbal medicines or Phytotherapy.

23.1- DRUG DISCOVERY AND DEVELOPMENT PROCESS

Drug discovery and drug development are important processes that help in the production of safe and effective medicines for the treatment of diseases. the process begins with the identification of disease-causing targets and the development of compounds that can interact with these targets. before a drug is approved for public use, it undergoes several stages of testing to evaluate its safety, efficacy, and quality. scientists, researchers, and regulatory authorities work together throughout this process to ensure that new medicines are beneficial and safe for human use**.**

23.1.1 STAGES OF DRUG DISCOVERY:

It is the initial stage of finding a new drug for any disease.There are following stages in the discovery of modern drugs;

1. Target Identification:

Scientists identify a specific molecule, such as a protein or enzyme, that is involved in causing a disease. This target is selected because it can be affected by a drug to treat the disease.

2. Target Validation

Researchers confirm that the identified target plays an important role in the disease process. Experiments are carried out to ensure that acting on the target can produce therapeutic effects.

3. Lead Compound Identification

Scientists screen thousands of natural or synthetic compounds to find substances that can interact effectively with the target molecule. These promising substances are called lead compounds.

4. Lead Optimization:

These lead compounds are modified to improve their effectiveness, reduce toxicity, and enhance the drug-like properties.

23.1.2 DRUG DEVELOPMENTAL PROCESS:

It is the process of testing and preparing the discovered drug for public use. There are following stages to develop a new drug;

1. Preclinical Research:

Compounds are tested in *vitro* (cell-based culture) and in *vivo* (animal) to measure safety, toxicity, and efficacy before human trials.

2. Clinical Trials:

Drugs are tested on humans to ensure safety.

Phase 1: Testing in a small group (20–100) of healthy volunteers are taken to determine safety and dosage.

Phase 2: Testing in a larger group (100–300) of patients to test the effectiveness and side effects of drugs.

Phase 3: Large-scale trials (1,000–3,000) are taken to confirm the efficacy and monitor their adverse reactions in patients.

3. FDA Drug Review:

Regulatory agencies (FDA or EMA) thoroughly examine all data from clinical research trials to approve or reject the drugs before supply to market.

4. Manufacturing and Marketing

Once approved, the drug is produced on a large scale in pharmaceutical industries and made available for medical use.

5. Post-Marketing Surveillance

Even after release, the drug is continuously monitored for long-term effects and rare side effects to ensure ongoing safety in the population.

23.2- CLASSES OF ANTIBIOTIC:

Antibiotics are drugs that treat bacterial infection by killing bacteria or stopping their growth. Antibiotics are classified primarily by their mechanism of action and chemical

structure. The major four classes are;

1. Penicillin
2. Tetracycline
3. Fluoroquinolones
4. Sulfonamides

23.2.1 Penicillin:

Penicillin is a group of bactericidal, antibiotics derived from *Penicillium* fungi, used to treat various bacterial infections by inhibiting bacterial cell wall synthesis. Alexander Fleming (1928), discover it, which is highly effective against gram-positive bacteria, including streptococcal and staphylococcal infections. Common side effects of penicillin antibiotics include nausea, vomiting, diarrhea, abdominal pain, and skin rashes.

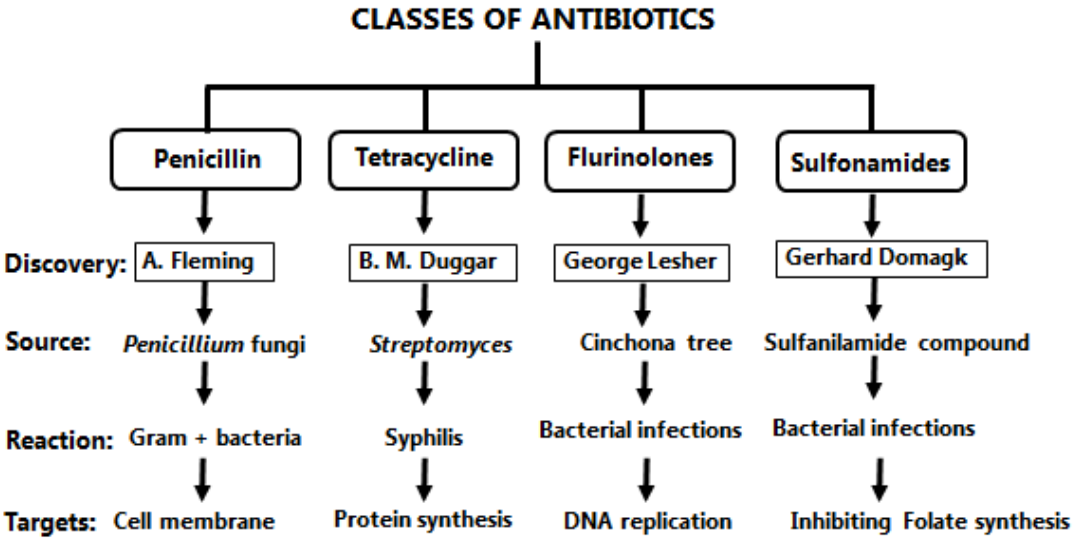


Fig: 23.1 Classification of Antibiotics

Mode of Penicillin Action:

Penicillin adopt following way to kill bacteria;

1. **Targeting Bacteria:** Penicillin target bacteria because animal cells lack the cell walls that are targeted by the drug.
2. **Cell Wall Inhibition:** It interrupt the cross-linking of peptidoglycan (murein) chains, stopping new cell wall synthesis of bacteria.
3. **Lysing bacterial Cell:** The bacterial cell wall can no longer maintain structural integrity and it may burst (lysis), after the attack of penicillin (kills rather than just inhibits growth).
4. **Resistance Mechanisms:** Some bacteria defend themselves by producing specific enzymes like beta-lactamases.

23.2.2 Tetracycline:

Tetracycline antibiotic is derived from soil-dwelling bacteria of the genus *Streptomyces*, It is a bacteriostatic oral antibiotic used to treat bacterial infections like chlamydia and syphilis. It inhibits bacterial protein synthesis. It should not be taken with dairy or iron supplements. Common side effects include nausea, vomiting, diarrhea, and sun sensitivity.

Mode of Tetracycline Action:

1. **Bacteriostatic Effect:** They stop bacteria from growing and multiplying rather than killing them outright.
2. **Gram-Negative/Positive Action:** They are effective against both Gram-positive and Gram-negative bacteria.
3. **Targeting Ribosomes:** Tetracycline enter bacterial cells and it reversibly bind to the 30S ribosomal subunit of its target cell.
4. **Blocking Protein Production:** By binding to the 30S ribosome, they prevent aminoacyl-t RNA from binding to the acceptor site, to inhibit translation and protein synthesis.

23.2.3. Fluoroquinolones:

Fluoroquinolones are synthetic, antibacterial agents derived as a byproduct of chloroquine (quinine) synthesis. Chloroquine is a synthetic drug derived from structural modification of quinine, a natural alkaloid found in the bark of the South American Cinchona tree. Fluoroquinolones was discovered by George Lesher (1962), are a class of bactericidal antibiotics that treat bacterial infections by inhibiting bacterial DNA replication. They are also associated with significant, rare adverse side effects, including tendon ruptures, peripheral neuropathy, drug resistance and cardiac issues.

Mode of Fluoroquinolones Action:

1. **Bactericidal Effect:** Fluoroquinolones trap the enzymes involved in DNA replication, creating a complex that causes double-strand DNA breaks. This prevents the bacteria from multiplying and forces them to die, so it classified as bactericidal (kills bacteria).
2. **Targeting Topois omerases:** Fluoroquinolones enter the bacterial cells and inhibit their Type II topoisomerases.
3. **Targeting Topoisomerase IV:** Primarily targeted in Gram-positive bacteria, it stops the separation of replicated DNA.
4. **Targeting DNA Gyrase:** Primarily targeted in Gram-negative bacteria, it stopping the process of DNA uncoiling during replication.

23.2.4 Sulfonamides:

Sulfonamides are synthetic pharmaceutical compounds derived from sulfanilamide, discovered by Gerhard Domagk in 1932. These are antimicrobial drugs that can treat

bacterial infections by inhibiting folate synthesis, primarily used for urinary tract infections and skin infections. It acts as competitive inhibitors and bacteriostatic antibiotics to inhibit bacterial synthesis of folic acid, which is essential for DNA synthesis and bacterial growth. These are often administered orally but have few side effects like rashes, nausea, photosensitivity and kidney damage.

Mode of Sulfonamides Action:

1. **Bacteriostatic Effect:** Sulfonamides do not kill bacteria immediately but inhibit their growth, allowing the body's immune system to destroy them.
2. **Selective Toxicity:** Sulfonamides are selectively toxic to bacteria because of how they handle folate. Bacteria must build this essential vitamin from scratch, whereas humans simply absorb it from food. This allows the drug to starve the bacteria of vital nutrients while leaving human cells untouched."
3. **Competitive Inhibition:** Sulfonamides bind to the enzyme responsible for creating folate, essentially taking para amino benzoic acid pathway (Folate metabolism).
4. **Combined Therapy:** They are often combined with other agents, like trimethoprim, to create a stronger bactericidal effect (killing bacteria).
5. **Broad Spectrum:** They are effective against both Gram-positive and Gram-negative bacteria, often used for urinary tract infections, acne, and respiratory infections.

23.3 ANTIVIRAL AND ANTIRETROVIRALS DRUGS

Antiviral drugs are used to treat viral infections. They prevent the growth and multiplication of viruses within the body. These medications can combat a wide range of viruses, including the hepatitis, influenza, and herpes viruses.

> All Antiretroviral are Antiviral but all Antiviral are not Antiretroviral

Antiretroviral drugs are designed to treat illnesses brought on by retroviruses, including HIV (Human Immunodeficiency Virus). They aid in immune system defense and stop HIV from proliferating. Retroviruses are viruses that replicate by using enzyme reverse transcriptase and RNA as their genetic material.

23.4- MONOCLONAL ANTIBODIES:

These are artificially laboratory-produced molecules that enhance the immune system's ability to fight against pathogens. Monoclonal antibodies target specific antigens, making them effective for treating cancer, autoimmune disorders, and infectious diseases.

23.4.2- ADVANTAGES OF MONOCLONAL ANTIBODIES

Monoclonal antibodies are integral to personalized medicine, enabling tailored treatment approaches based on individual patient profiles. Continued innovation in

these antibodies in design and production is critical to improve its effectiveness of therapies, ensuring they are better suited to each patient's unique biology.

Monoclonal antibodies have significant advantages as;

i. As compared to traditional drugs, they offer lower toxicity, potential for long-lasting effects.
ii. These have least side effects as compared to conventional chemotherapeutics.
iii. These have high specificity, binding to a single target with minimal variability.
iv. They provide targeted therapy in cancer, reducing damage to healthy tissues.
v. These are easily to prepare and used for research purpose.
vi. They reduced the risk of drug interactions.

These technologies enable the simultaneous testing of thousands of potential antibodies, reducing development timelines and enabling faster responses to urgent health challenges like infectious diseases and cancer.

EXERCISE

SECTION 1: MULTIPLE CHOICE QUESTIONS

1. The first step in drugs discovery is _____.
   (a) Lead optimization (b) Preclinical Testing
   (c) Target identification (d) Hit identification
2. Testing in a larger group about 100–300 patients to test the effectiveness and side effects of drugs indicate _____
   (a) Phase 1 (b) Phase 2
   (c) Phase 3 (d) Phase 4
3. Penicillin is highly effective against _____
   (a) Gram-positive bacteria (b) Gram-negative bacteria
   (c) *Streptomyces* (d) *Penicillium notatum*
4. The _____ antibiotic derived from soil-dwelling bacteria of the genus Streptomyces,
   (a) Penicillin (b) Fluoroquinolones
   (c) Sulfonamides (d) Tetracycline
5. The _____drugs are synthetic compounds derived from sulfanilamide,
   (a) Penicillin (b) Fluoroquinolones
   (c) Sulfonamides (d) Tetracycline
6. Monoclonal antibodies are considered highly affect because
   (a) have broad spectrum action (b) are highly specific to antigens
   (c) do not bind to any target (d) are non-biological in nature

SECTION 2: SHORT QUESTIONS

1. Briefly describe Stages of Drug Developmental Process?
2. Write side effects of Penicillin?
3. Write a mode of Tetracycline action?
4. Differentiate between antiviral and antiretroviral drugs?
5. What are the advantages of monoclonal antibodies?

SECTION 3: LONG QUESTIONS

1. Describe the essential steps for the drug discovery and developmental process?
2. Explain different type of antibiotics. Describe their mode of action.
3. Describe the Monoclonal Antibodies and their advantages?

INQUISITIVE QUESTIONS

1. Predict the challenges the scientists may face during drug development process and propose possible solutions.
2. Assess the possible consequences of the misuse and overuse of antibiotics on human health.

Chapter 24

EVOLUTION

STUDENTS'LEARNING OUTCOMES

After studying this chapter, the students will be able to:

- Explain that evolution happens due to variation in organisms and the selection pressures that organisms face.
- Analyze the evidence of evolution that comes from molecular biology and that is provided by biogeography.
- Differentiate between convergent and divergent evolution on the basis of inheritance of the homologous and analogous structures.
- Describe the endosymbiotic theory about the mechanism of evolution of eukaryotes from prokaryotes.
- Describe the theory of inheritance of acquired characters, as proposed by Lamarck with example of giraffe neck.
- State the drawbacks in Lamarckism.

This chapter would deliver you the concept of evolution of life through the theories put forwarded by different scientists. You will also analytically review the major evidence that form the basis of the most accepted theory of evolution.

According to the theory of **Creationism** ("theory of intelligent design"), all organisms were created as such as they exist today. Creationists claim that each type of organism was created individually and did not go through any change throughout the history.

24.1- The Concept of Evolution

Evolution is the gradual change in the genetic makeup (allele frequencies) of a population over successive generations, which may lead to the formation of new species. In its earliest form, the concept of evolution had the following basic speculations:

Early people also believed that living creatures can also develop from non-living matter. It was called idea of **spontaneous generation**. With the progress of science, the experiments disproved spontaneous generation but such experiments could not negate creationism.

- The universe and organisms did not always exist in their present form;
- Today's organisms were not created at once; and
- They are the result of innumerable changes from the lower to the higher form.

Evolution takes place due to variations in organisms and natural selection. According to this idea, organisms with beneficial traits are selected to produce next

generations. On the other hand, the individuals with disadvantageous traits cannot produce next generations. It results in the increase in the percentage of individuals with better traits in populations.

24.2- Theory of Evolution - Darwinism

Charles Darwin was an English scientists. In 1831, he travelled as a naturalist on scientific expedition on His Majesty's Ship called the HMS Beagle. He made a 5-year voyage around the world.

Figure 24.1: Main stops in the route of HMS Beagle

Figure 24.2: Charles Darwin

Darwin's Observations

During the long voyage, Darwin visited tropical rainforests and other new habitats where he saw great diversity in plants and animals that he had never seen before. He also found rocks containing fossils. These observations suggested that continents and oceans had changed dramatically over time and continue to change. He dug up fossils of gigantic extinct mammals and found that organisms looked very different in the past. It suggested that living things change over time.

When he visited the Galápagos Islands (a group of 16 small islands), he noticed that the plants and animals on different islands were very different. For example, the giant tortoises on one island had saddle-shaped shells, while those on another island had dome-shaped shells. Similarly, many plants and animals resembled more with the plants and animals of South America, rather than with other islands. Moreover, different species of the common birds i.e., finches had different beaks. He found that differences in the sizes and shapes of beaks were actually adaptations for feeding on completely different kinds of food at different islands.

Darwin thought over these adaptations and tried to develop an explanation for the distribution of finches species among the islands. He thought that the origin of new species and adaptations are closely related processes. A new species would arise from an ancestral form by the gradual accumulation of adaptations to different

environment, separated from original habitat. Over many generations, the two populations could become dissimilar enough to be designated as separate species.

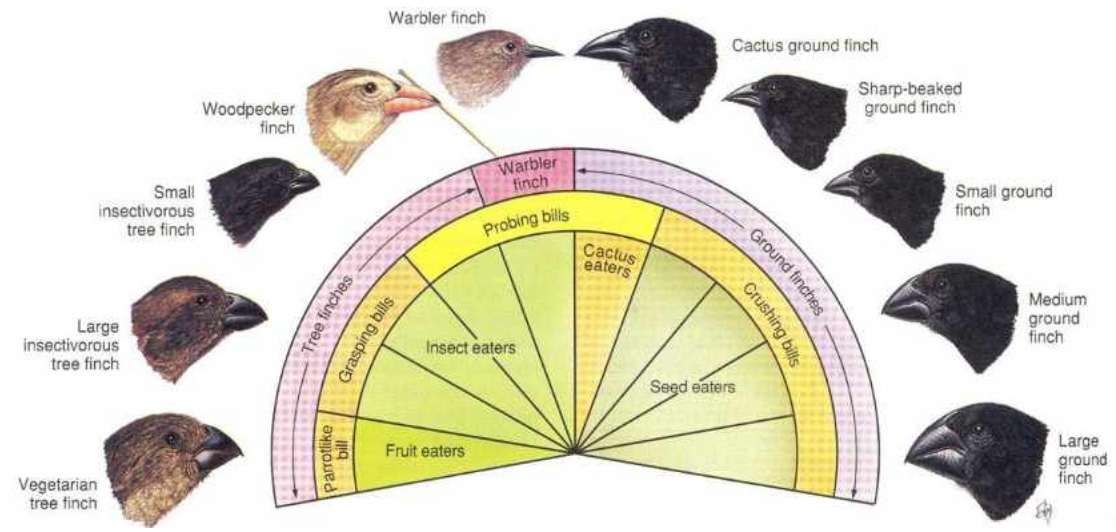


Figure 24.3: Adaptations in beak size and shape in different finches of Galapagos Islands

24.2.1- Mechanism of Evolution - Natural Selection

The founder of the modern theory of evolution was **Charles Darwin** (1809 – 1882). In 1859, he published his book **"On the Origin of Species"**. In this book, he proposed that evolution takes place due to variations in organisms through natural selection. The essence of his theory is as follows:

1. **Overproduction:** All organisms have a far greater reproductive potential than is ever realized. For example, a female sea star releases about 1 million eggs each season. What if all of these eggs were fertilized and developed to reproductive adults by the following year? A half million female sea stars (half of the million eggs would produce females and half would produce males), each producing another million eggs, repeated over just a few generations would soon fill up the oceans!
2. **Variations:** Genetic variations arise by random mutations. Seldom are any two individuals exactly alike. These genetic variations may be advantageous, harmful or neutral (neither helpful nor harmful). Inherited variations can be passed on to offspring.

Alfred R. Wallace (1823 – 1913) was a British naturalist and biologist who also developed his own theory on the mechanism of evolution. In 1957, he wrote a letter to Darwin. In Wallace's letter, the proposed mechanism of evolution was very similar to the theory of Darwin. When both Darwin's and Wallace's theories were presented to the Linnaean Society in 1858, Wallace agreed to attribute the theory to Darwin. He convinced Darwin to publish his book "On the Origin of Species".

3. **Struggle for survival:** Because many more offspring are produced than resources can support, individuals struggle for survival. The individuals with disadvantageous variations die or cannot reproduce. Individuals with advantageous variations remain successful in reproduction. Such variations are called adaptive traits.
4. **Inheritance of adaptive traits:** Adaptive traits are inherited to next generations. Because organisms with disadvantageous traits are less likely to reproduce, such traits become less frequent in a population and eventually are eliminated.

24.2.2- Neo-Darwinism

In the early 20th century, the Darwinism was integrated with modern genetics to develop Neo-Darwinism. According to Neo-Darwinism, a more precise definition of evolution is "a change in the frequency of alleles in a population or in the total genetic makeup of a population (the gene pool)". Neo-Darwinism is based on Hardy-Weinberg Theorem.

Hardy-Weinberg Theorem

In 1908, English mathematician **Godfrey Hardy** and German physician **Wilhelm Weinberg** presented Hardy-Weinberg theorem. It states that the frequencies of alleles and genotypes in a large, randomly mating population remain constant from generation to generation, if no evolutionary forces are acting. According to Hardy-Weinber theorem, any of the following conditions is not followed, evolution takes place.

1. The population size must be large.
2. Mating within the population must be random. Every individual must have an equal chance of mating with any other individual.
3. Individuals must not migrate into or out of the population.
4. Mutations must not occur. If they do, mutations from the wild-type allele to mutant allele must be equal to mutations from the mutant form back to the wild type.
5. All genotypes must have an equal chance of survival and reproduction.

For Information

Gene pool means the total number of all genes for a trait in a population. Example: If a population has 1000 genes for flower colour, this is its gene pool.

Allele (gene) frequency is the number or proportion of a specific allele in the population. Example: If 600 out of 1000 genes are for red colour, the allele frequency of red is 0.6 (60%).

Genotype frequency is the number or proportion of individuals with a particular genotype in a population. Example: If 40 out of 100 plants are RR, the genotype frequency of RR is 0.4 (40%).

Factors that can change the allelic frequencies

The following are the factors due to which the Hardy-Weinberg assumptions are not met and changes in allelic frequencies occur from one generation to the next.

1- Genetic Drift or Neutral Selection

When chance events influence the frequencies of genes in populations, such change is called genetic drift. Unlike natural selection, these changes occur by chance - not fitness - and are most obvious in small populations. Genetic drift is often called neutral selection. For example, a population of rabbits is reduced from 100 to 10 by a fire. The survivors happen to be mostly brown, not because they were better adapted, but by chance. The resulting population will have lower genetic diversity.

2- Gene Flow

The Hardy-Weinberg theorem assumes that no individuals enter a population from the outside (immigrate) and that no individuals leave a population (emigrate). The entry of individuals from another population can introduce new mutations, instantly changing gene frequencies (evolution). Changes in gene frequency due to migration of individuals are called gene flow.

3- Mutation

The Hardy-Weinberg theorem assumes that no mutations occur or that mutational equilibrium exists. Mutations, however, are a fact of life. Most importantly, mutations are the origin of all new genes and a source of variations that may prove adaptive for an organism. Mutations also make sure that variations will be present that allow a group to survive environmental shocks more successfully.

The effects of mutations vary. Most are deleterious. Some may be neutral or harmful in one environment and help an organism survive in another environment. Mutational equilibrium exists when a mutation from the wild-type allele to a mutant form is balanced by mutation from the mutant back to the wild type. This has the same effect on allelic frequency as if no mutation occurred. However, mutational equilibrium rarely exists. Mutation pressure is a measure of the tendency for gene frequencies to change through mutation.

24.3- Evidences of Evolution

24.3.1- Evidence from Biogeography

Biogeography (study of the geographic distribution of plants and animals) shows that life-forms in different parts of the world have distinctive evolutionary histories. For example;

- Similar groups of organisms can live in places separated by vast barriers. Obvious similarities suggest a common ancestry, but similarly obvious differences result from millions of years of independent evolution. For example, African lion is very much similar to mountain lion of America. Their similarities suggest a common ancestry. However, obvious differences result from millions of years of independent evolution.

- Organisms separated by geographical barriers, are often very different in spite of similar environments. For example, many animals living in Australia and Tasmania are very different from animals in any other part of the world. Many species of kangaroos are the major herbivores of Australia and Tasmania.
- Islands often have relatively few but unique species due to island colonization and subsequent evolutionary events.

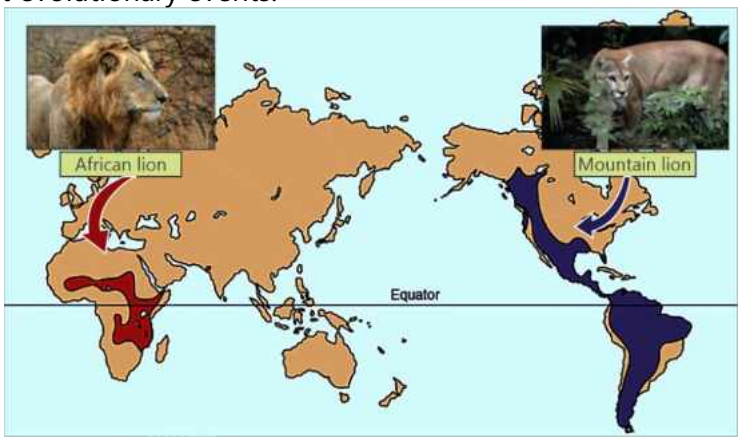


Figure 24.4: Distribution of lions found in Africa and North and South America

24.3.2- Evidence from Comparative Anatomy

Comparative anatomists study the structure of fossils as well as living organisms, looking for similarities that could indicate evolutionarily relationships. This study reveals two types of evolution.

a- Divergent evolution: Body parts that are similar in structure but different in function are called **homologous** structures and this phenomenon is called **homology**. Homologous structures reveal divergent evolution which means that the organisms with homologous structures have a common ancestor and evolved into different types and used their structures for different functions. For example, vertebrate appendages have a common arrangement of similar bones, even though the function of the

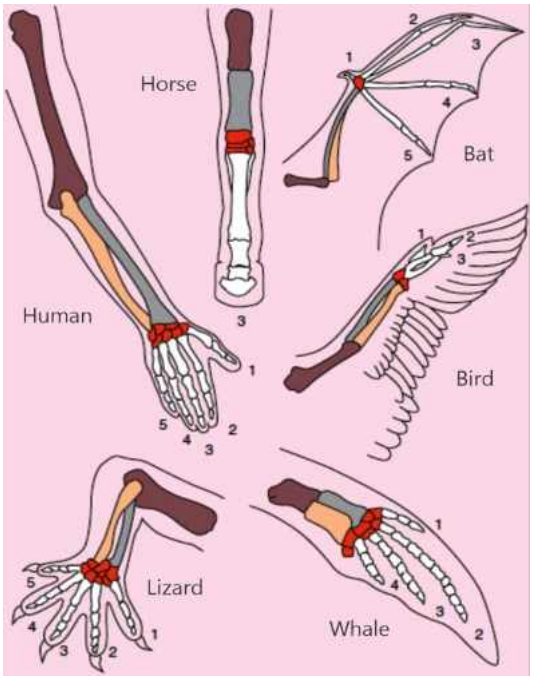


Figure 24.5: Homology in vertebrates forelimbs

appendages may vary i.e., flights (birds and bats), swimming (whales and dolphins), running (horses), climbing (arboreal lizard), or swinging from tree branches.

b- Convergent evolution: The organs which are similar in function but differ in structure are called analogous organs. Such structures present in different organisms indicate convergent evolution i.e., the evolution in which different types of organisms have been evolved from different ancestors at a common habitat. For example, the wing of a bird and the wing of an insect are both adaptations for flight, but they are not homologous. Any similarities simply reflect the fact that, to fly, an animal must have a broad, flat gliding surface.

Vestigial organs: Organisms often retain structures that have lost their usefulness. These structures are often poorly developed and are called vestigial structures. For example, some snakes like boa constrictors and pythons have minute remnants of hindlimb (pelvic) bones left over from appendages of their reptilian ancestors. Such remnants of once useful structures are clear indications of change i.e., evolution.

Vestigial structures in humans

There are about 90 vestigial structures present in human body e.g.,

- Vermiform appendix is a vestige of the cecum.
- Coccyx or tailbone is the remnant of tail.
- The wisdom teeth are vestigial third molars that human ancestors used to help in grinding down plant tissue.
- Ear muscles are minimally developed and non-functional, but some people are able to move their ears in various directions.

24.3.3- Evidence from Molecular Biology

Recently, molecular biology has yielded a wealth of information on evolutionary relationships. Just as animals can have homologous structures, they may also have homologous biochemicals. Ultimately, structure and function are based on the genetic blueprint (DNA molecule) found in all living animals.

Related animals have similar DNA derived from their common ancestor. Because DNA carries the codes for proteins that make up each animal, related animals have similar proteins. Scientists can extract and analyse the structure of DNA and proteins of different organisms. By looking for dissimilarities in the structure of related proteins and DNA, and by assuming relatively constant mutation rates, scientists can estimate the elapsed time since evolution from a common ancestral molecule.

How microorganisms have evolved resistance to antibiotics?

Antibiotic resistance means that bacteria are able to survive under the exposure of an antibiotic. It may be due to mutations in bacterial genes. A gene for antibiotic resistance which had evolved via natural selection passes to next generations of bacteria.

24.4- Evolution From Prokaryotes To Eukaryotes

As we know that the earliest organisms on the Earth were prokaryotes who originated in a very unstable environment from the reactions of organic molecules about 3.5 billion years ago. About 2 billion years ago, eukaryotes evolved from prokaryotes. The "Endosymbiotic Theory" is the most acceptable theory for the evolution of prokaryotes into eukaryotes.

Endosymbiotic Theory

This theory was proposed by **Lynn Margulis**. According to this theory, large prokaryotes engulfed small prokaryotes. The small prokaryotes were not digested by the larger ones. Instead, they lived within the large cells and evolved into organelles.

The large and small prokaryotes formed a symbiotic relationship in which both cells benefited. Some of the small prokaryotes were able to do aerobic respiration. They supplied energy not only to themselves but also to the large cell. They became the mitochondria of eukaryotic cells. Other small cells were able to use sunlight to make food. They shared the food with the large cell. They became the chloroplasts of eukaryotic cells. Similarly, flagella may have derived through the ingestion of prokaryotes similar to spirochetes.

Another theory called "Membrane Invagination Theory" also explains the evolution of eukaryotes. According to this theory, the cell membrane of prokaryotic cell invaginated in the cytoplasm. It gave rise to some membranous structures including nucleus and endoplasmic reticulum inside prokaryotic cell. It resulted in the formation of a prokaryotic cell

Evidence for the endosymbiotic theory

- Mitochondria (and chloroplasts) are of the same size as prokaryotic cells.
- Mitochondria (and chloroplasts) have their own DNA. Their DNA is organized in a circular chromosome like a prokaryotic chromosome.
- Mitochondria (and chloroplasts) can reproduce by binary fission like prokaryotes. In contrast, eukaryotic cells reproduce by mitosis.
- The membrane composition of mitochondria (and chloroplasts) is more similar to prokaryotic membranes than to eukaryotic membranes.

Eukaryotic cells, evolved by endosymbiosis, were powerful and efficient. Their power and efficiency enabled them to evolve new characteristics e.g., multicellularity, cell specialization, and large size. They were the key to the spectacular diversity of animals, plants, and fungi that populate our world today.

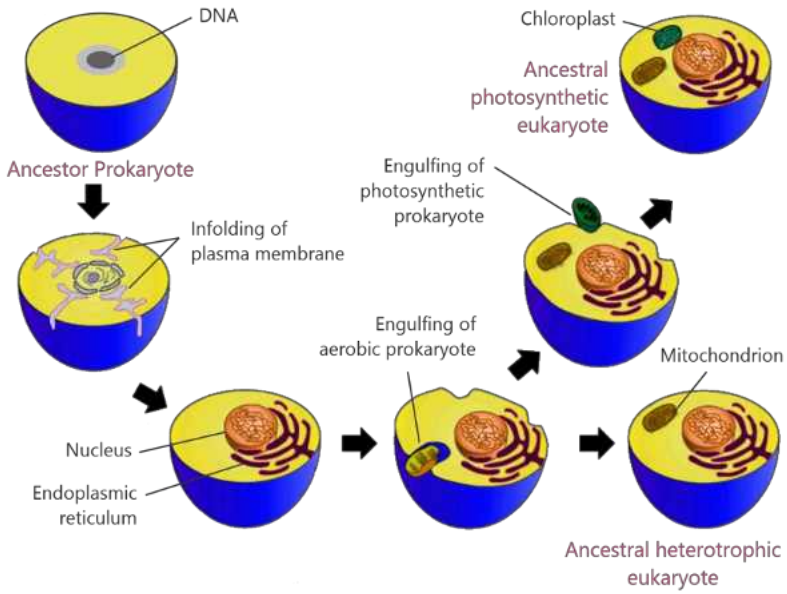


Figure 24.6: Evolution of Eukaryote

24.5- LAMARCKISM

The French naturalist **Jean-Baptiste Lamarck** (1744 - 1829) was an early proponent of the idea that evolution. In 1809, he published a book 'Philosophie Zooloqique' (Zoological Philosophy). In this book, he presented the idea of progression (change) known as Lamarckism.

According to Lamarckism, organisms changed from simplest to complex. He claimed that changes occur in organisms when they are adapted to environment. The mechanism of evolution proposed by Lamarck can be explained as under;

1- Use and disuse of organs

Lamarck believed that when an organism faces changes in its environment, it begins to use some body parts / organs more or disuses some organs. He believed that the organs which are used more frequently are developed and become strong while the organs which are not properly used are deteriorated or become weak. He named such changes in body parts, which occur due to long term use or disuse, as "acquired characters".

Figure 24.7: J. B. Lamarck

2- Inheritance of acquired characters

Lamarck believed that the characters acquired during the lifetime of an organism were passed on to the offspring of that organism. When acquired characters are inherited generations to

generations, they ultimately cause modifications in organism to better "cope" the changes in the environment.

Example of Lamarckism: Evolution of giraffe neck

The evolution of a giraffe's neck is often used as the example to prove Lamarckism. The ancestors of giraffe did not have long necks. They used to eat grass or the leaves of small plants. Changes occurred in the environment and small plants were no more available to them. In the new situation, some individuals of the population began stretching their necks to reach the leaves of tall trees as alternative. When they constantly did so, their necks became strong and long during their lifetime. This was an acquired character. When these individuals reproduced, this acquired characters was inherited to their next generation i.e., new individuals were born with slightly longer necks. When these individuals grew up, they continued the same practice of stretching their necks. In this way, their necks became even more stronger and longer. This gave rise to an acquired character. When this acquired character was inherited to many generations, the generation with long necks evolved.

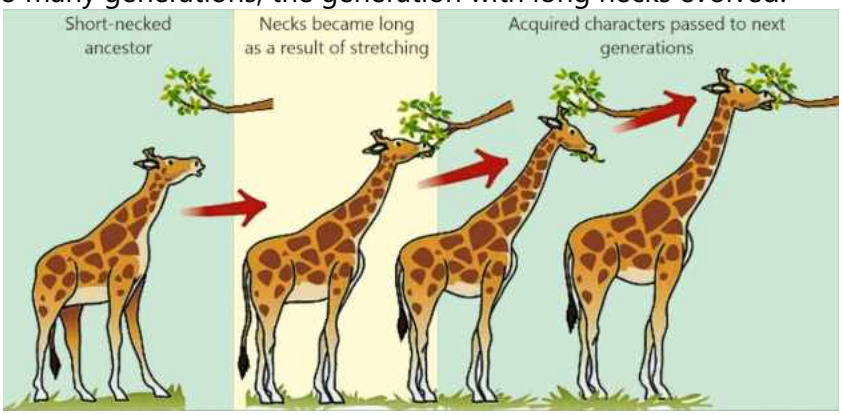


Figure 24.8: Evolution of giraffe neck – according to Lamarckism

24.5.1- Drawbacks of Lamarckism

The scientists did not accept the ideas of change as proposed by Lamarck. At the times when Lamarckism was proposed, there was no available evidence that could explain the mechanism of evolution. Thus, Lamarck was criticized more for advocating ideas of evolutionary change than for the mechanism he proposed for that change. After his times, when scientists discovered the inheritance mehcanisms, they disproved Lamarckism on the following bases.

Initiating and Planning
Hypothesize whether Lamarck was criticized in his day for advocating the ideas of evolution or for the mechanism he proposed.

1. The acquired characters have no genetic basis and therefore cannot be inherited to the next generations.
2. Organs are not modified by the wish or requirements of an organism.

EXERCISE

SECTION 1: MULTIPLE CHOICE QUESTIONS

1. Who developed a theory of natural selection essentially identical to Darwin's?
   (a) Hardy-Weinberg (b) Alfred Wallace
   (c) Malthus (d) Lyell
2. Which statement is incorrect?
   (a) Homologous organs are functionally different but structurally alike
   (b) Analogous organs are functionally different but structurally alike
   (c) Examples of analogous structures are arms of a man, forelimb of a cat, flipper of a whale
   (d) Examples of homologous structures are wings of bats, birds and insects
3. Which of the following is an example of homologous organs?
   (a) Wings of bird and insect
   (b) Forelimbs of human and whale
   (c) Fin of fish and wing of bird
   (d) Eye of octopus and human
4. According to the theory of natural selection, variations arise due to;
   (a) Mutations (b) Use and disuse of organs
   (d) Selective breeding (d) Splitting of population
5. Which condition is NOT required for Hardy-Weinberg equilibrium?
   (a) Random mating (b) No mutation
   (c) Natural selection (d) Large population size
6. Genetic drift is the change in gene frequency due to;
   (a) Chance events (b) Natural selection
   (d) Emigration of individuals (d) Immigration of individuals
7. Which of the following ideas is common to both Lamarckism and Darwinism?
   (a) Mutations are the source of genetic variations
   (b) Adaptations enable individuals to cope with environmental challenges
   (c) Use and disuse of organs results in the development of acquired characters
   (d) Acquired characters are inherited to next generations
8. If a massive fire kills 90% of a population and survivors remain by luck. Such a change in allelic frequency will be called;
   (a) Natural Selection (b) Genetic Drift
   (c) Gene Flow (d) Mutation
9. Changes in gene frequency due to migration of individuals are called;
   (a) Genetic drift (b) Mutation
   (c) Gene flow (d) Genetic recombination

10. What may be the best evidence for the endosymbiotic evolution of eukaryotes?
    (a) Presence of a nucleus in eukaryotes
    (b) Circular DNA and 70S ribosomes in mitochondria
    (c) Single-layered membranes around mitochondria
    (d) Large volume of eukaryotic cells

SECTION 2: SHORT QUESTIONS

1. What is the concept of evolution?
2. How does homology provide an evidence of divergent evolution?
3. How analogous organs represent convergent evolution?
4. What is Hardy-Weinberg theorem?
5. What are the factors that change allele frequencies?
6. Justify Lamarck as an early proponent of evolution.
7. Describe the drawbacks of Lamarckism.
8. Differentiate between;
   - Lamarckism and Darwinism
   - Homologous and analogous
   - Natural selection and neutral selection

SECTION 3: LONG QUESTIONS

1. Explain that evolution takes place due to variations in organisms and natural selection.
2. Explain the theory of natural selection as proposed by Darwin.
3. Explain how biogeography provides evidence for evolution.
4. Describe the evidences of evolution that come from comparative anatomy.
5. Differentiate between convergent and divergent evolution on the basis of inheritance of the homologous and analogous structures.
6. Describe the evidences of evolution that come from molecular biology.
7. Outline the steps of the evolution of the giraffe, as illustrated in Lamarckism.
8. Describe the endosymbiont theory about the evolution of eukaryotes from prokaryotes.
9. Describe the theory of inheritance of acquired characters, as proposed by Lamarck.

INQUISITIVE QUESTIONS

1. Interpret different homologous and analogous structures through observation in plants.
2. Hypothesize whether Lamarck was criticized in his day for advocating the ideas of evolution or for the mechanism he proposed.

3. Justify, on the grounds that both Wallace's and Darwin's papers were published in the Journal of the proceedings of the Linnaean Society, why the theory was attributed to Darwin.
4. Describe and analyse examples of technology that have extended or modified the scientific understanding of evolution (e.g., the contribution of radiometric dating to the palaeontological analysis of fossils).

Chapter 25

ECOLOGY

STUDENTS'LEARNING OUTCOMES

After studying this chapter, the students will be able to:

- Define species, population, community and ecosystem.
- Define biogeochemical cycles and locate the primary reservoirs of the chemicals in these cycles.
- Describe the water cycle in detail.
- Discuss nitrogen cycle in detail.
- Discuss the loss of energy between trophic levels.
- Describe characteristics of a population, such as growth, density, distribution, carrying capacity, minimum/viable size.
- Explain the effect of growth of human population on the ecosystem.
- Explain the greenhouse effect with examples of gases that exhibit this behavior.
- Describe the harmful effects of greenhouse gases on the environment.
- Describe four important ecosystems of Pakistan.

All organisms have certain requirements for life. They fulfill the requirements by interacting with other organisms and with the physical environment. Ecology is the study of the relationships of organisms with other organisms and the environment. In this chapter, you would get learning on the basic concepts of environment and the cycling of materials and energy between the environment and organisms.

25.1- Levels of Ecological Organization

To understand how life works, we must look through different "levels" on which natural world is organized into layers of increasing complexity.

1. Species: The Unit of Life

A species is the basic unit of biological classification. It refers to "a group of organisms that share similar physical and genetic characteristics and, most importantly, can interbreed to produce fertile offspring". For example, The Indus River Dolphin is a distinct species. All Indus dolphins can breed with one another, but they cannot breed with other types of dolphins to produce fertile young.

2. Population

A population is a group of individuals belonging to the same species living in the same geographic area at the same time. A species refers to a "type" of organisms globally, while a population refers to a specific group in a specific location. For example, all the Markhors living in the Chitral Gol National Park form a single

population. If we consider Markhors across all of Pakistan, we are referring to the species; if we consider only about those in Chitral, we are referring to a population.

3. Community

In nature, no population lives alone. A community consists of all the different populations of various species living and interacting in the same area. This includes plants, animals, fungi, and bacteria. For example, a Mangrove Forest in Karachi refers to a community that includes the Mangrove trees, the mudskipper fish, the crabs, and the various shorebirds. They all share the same space and interact with one another (e.g., birds feed on fish).

4. Ecosystem

Communities live in ecosystems. An ecosystem consists of the biotic (living) components and the abiotic (non-living) components of the environment, such as sunlight, soil, water, and air. In an ecosystem, energy flows unidirectionally while the nutrients cycle between the living and the non-living parts.

For example, the Thar Desert is a vast ecosystem. It not only contains the snakes, blackbucks, and shrubs (the community); but also has the baking heat, the shifting sand dunes, and the rare rainfall (the abiotic factors) that influence the lifestyle of the organisms living in it.

5. Biome

A biome is a large, distinct geographical region characterized by its specific climate, soil altitude, and types of plants and animals which are adapted to those conditions.

In Pakistan, the diverse geography makes several biomes. For example; Coniferous Forest Biome is located in the high-altitude areas of Murree and Swat, where cold winters and pine trees dominate. Desert Biome is located in the Thal and Thar regions, characterized by low rainfall and specialized "succulent" plants like cacti. Similarly, Freshwater Biomes include the massive Indus River system.

6. Biosphere

The biosphere is the highest and most inclusive level of ecological organization. It is the global sum of all biomes. It refers to every part of the Earth where life exists - from the deepest trenches of the Arabian Sea to the highest peaks of the Himalayas, and even several kilometers up into the atmosphere.

Tidbit

The biosphere is often called the "Global Ecosystem". It is a thin "envelope" around the Earth where the lithosphere (land), hydrosphere (water), and atmosphere (air) interact to support life.

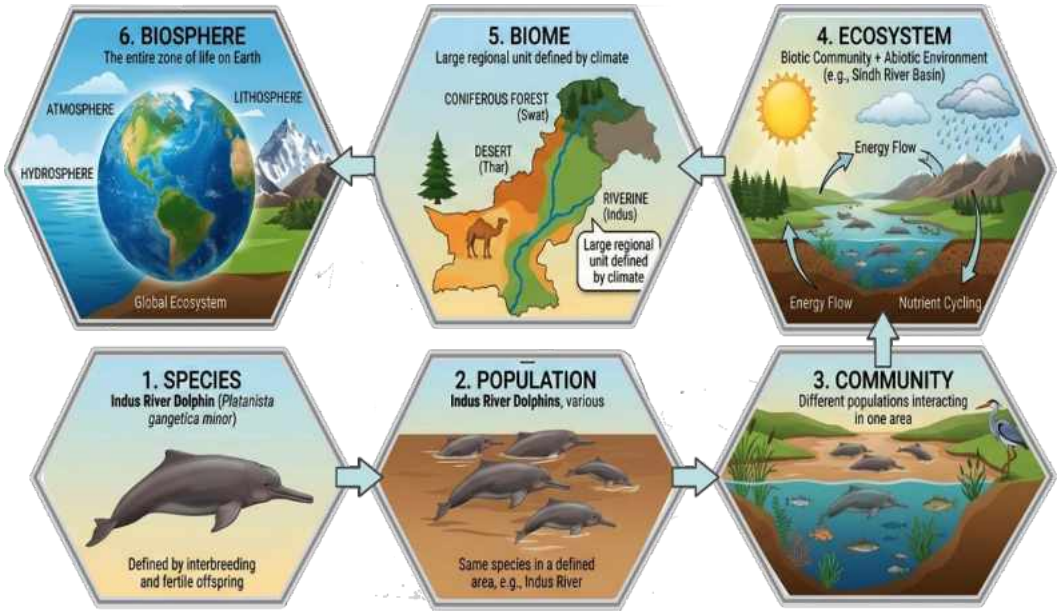


Figure 25.1: Levels of ecological organization

25.2- Biogeochemical Cycles

Biogeochemcial cycles are the cyclic movements of chemical elements – such as carbon, nitrogen, oxygen, phosphorous – between the living (biotic) and non-living (abiotic) components of ecosystem.

Primary Reservoirs of Biogeochemical Cycles

A primary reservoir ("sink" or "pool") is a massive component of the biosphere where the element of a biogeochemical cycle is stored for long periods of time. On the basis of primary reservoir, the biogeochemical cycles are classified as;

- **Gaseous cycles**, in which the reservoir is the air or the oceans (via evaporation). Gaseous cycles include nitrogen cycle, oxygen cycle, carbon cycle, and water cycle.
- **Sedimentary cycles**, in which the reservoir is Earth's crust. Sedimentary cycles include phosphorus cycle, sulfur cycle, and calcium cycle etc.

25.2.1- The Water Cycle

The water cycle (hydrological cycle) is the continuous movement of water between abiotic components (air, soil, and sea) and biotic components.

Tidbit
In a country like Pakistan, where our agriculture and economy depend entirely on the Indus River system, the water cycle is the literal lifeline of the nation.

Abiotic Loop

1. Evaporation: The sun powers the entire water cycle. Solar radiation heats the surface water in oceans and lakes. It provides enough energy for water molecules to evaporate in the form of vapors. About 86% of global evaporation occurs from the oceans. In Pakistan, the Arabian Sea serves as the massive primary source of vapor that eventually travels inland.

2. Condensation: As water vapors rise into the atmosphere, they loses heat. When the air becomes cold enough, the vapors turn back into liquid droplets or ice crystals. These droplets cling to dust particles in the air to form clouds.

3. Precipitation: When the water droplets in clouds become too heavy, they fall due to gravity. Depending on the temperature, water falls as rain, snow, or sleet (a rain mingled with snow or hail).

4. Infiltration and Runoff: Once water hits the land, it takes one of two main paths to complete the cycle:

- **Infiltration:** Water seeps into the soil (percolation). It moves downward to reach the water table. This stores water underground as groundwater.
- **Surface Runoff:** It is the unconfined flow of water over the land surface. It occurs when water can no longer infiltrate into the soil. The surface runoff water flows into streams, rivers, and eventually to the oceans.

Tidbit
In Pakistan, surface runoff feeds the Indus River, while infiltration recharges the aquifers, we use for drinking water.

Biotic Loop

1. Water Entry into the Biotic Component: Water enters biotic components primarily through two methods. Terrestrial plants use root hairs to absorb water from the soil and use it for photosynthesis and as a structural support (turgor pressure). Animals take in water by drinking directly from freshwater sources and by eating food.

2. Water Exit from the Biotic Component: Once water has served its purpose inside an organism. it returns into the abiotic environment. **Transpiration** in plants is the major exit point for water. Plants release water vapors in atmosphere through the stomata present in their leaves. **Respiration** in plants and animals also produces water as a byproduct. This "metabolic water" is exhaled as vapors. In **excretion** and **egestion**, animals also release water through urine and feces, returning it to the soil or water bodies. During perspiration (sweating), animals release water through skin pores, which evaporates to the atmosphere. **Decomposition** of organisms' dead bodies also releases water back into the soil or atmosphere.

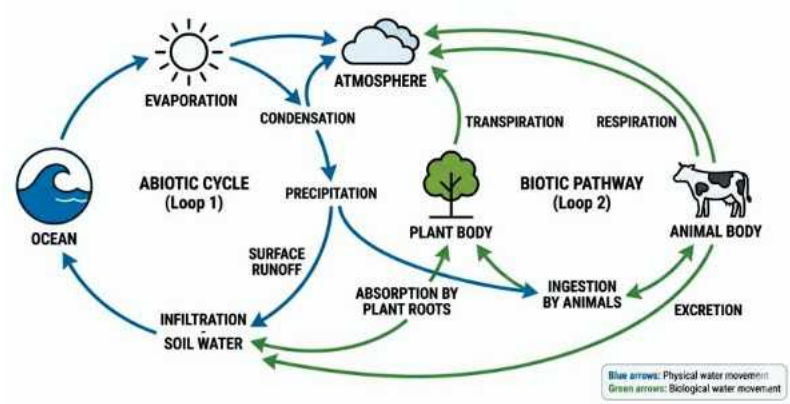


Figure 25.2: The steps of water cycle

25.2.2- The Nitrogen Cycle

The Nitrogen Cycle is a complex process by which nitrogen circulates between the biotic and abiotic components of the ecosystem. It involves the transformation of atmospheric nitrogen (N_2) into compounds that can be used by living organisms, and the subsequent return of N_2 to the atmosphere.

Tidbit
While the water cycle moves a molecule we can notice, the Nitrogen Cycle deals with an invisible gas that makes up about 78% of our atmosphere.

The Cyclic Path of Nitrogen

The nitrogen cycle moves in a continuous loop, passing through the following five essential steps:

1. Nitrogen Fixation (Entry into the Soil)

The atmosphere is 78% nitrogen gas (N_2). Its atoms are joined by a strong triple bond. Plants cannot break these bonds. Nitrogen fixation is the process of converting N_2 into inorganic compounds like ammonia (NH_3). It is done in three ways;

i. **Biological Fixation:** More than 90% of all nitrogen fixation is done by soil microorganisms. There are two kinds of nitrogen-fixing microorganisms: (i) free-living (non-symbiotic) organisms (e.g., cyanobacteria or blue-green algae, *Azotobacter*, and *Clostridium)* and (ii) mutualistic (symbiotic) bacteria (e.g., *Rhizobium*) present in the root nodules of many pulses (legumes) plants like Chana (Chickpeas) or Moong (Mung beans). These nitrogen-fixing microorganisms "fix" nitrogen directly from the air in the soil and convert it to ammonia.

ii. **Atmospheric Fixation:** During rains and storms, lightning provides the massive energy needed to combine atmospheric nitrogen with oxygen. So, N_2 is converted int nitrogen oxides and then into nitrates. Only a small amount (5-10%) of nitrogen is fixed in this way.

iii. **Industrial fixation:** The synthesis of nitrogen containing fertilizers is called industrial fixation. In this process, atmospheric nitrogen and hydrogen are combined to produce ammonia.

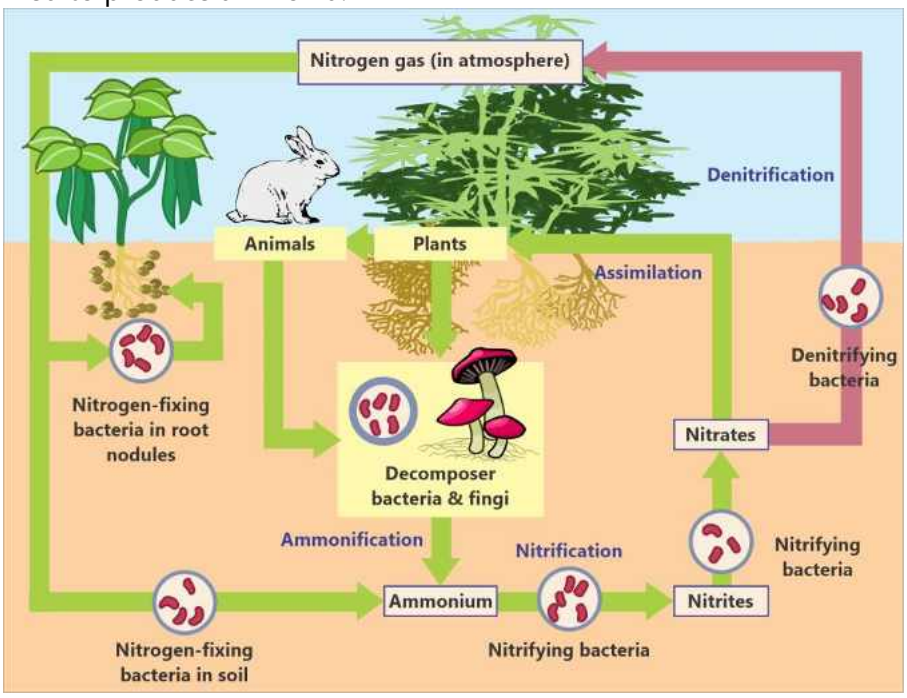


Figure 25.3: The nitrogen cycle

2. Nitrification (Conversion to Plant Food)

Ammonia is often toxic or difficult for plants to use in large amounts. Soil bacteria (such as *Nitrosomonas* and *Nitrobacter*) act as converters. *Nitrosomonas* convert ammonia to nitrite (NO_2^-). *Nitrobacter* convert this nitrite to Nitrate (NO_3^-). Nitrates are easily absorbed by plants.

3. Assimilation (Entry into the Biotic World)

Assimilation is the process in which plants and microbes absorb nitrates from the soil and use them to make proteins, DNA and chlorophyll. Animals assimilate nitrogen by consuming plants and other animals.

4. Ammonification (The Recycling Step)

When plants and animals die, or when animals release nitrogenous waste, **decomposers** (fungi and bacteria) break down the complex proteins. They convert the organic nitrogen back into Ammonia.

5. Denitrification (Exit to the Atmosphere)

To keep the cycle balanced, nitrogen must eventually return to the air. In

waterlogged soils, denitrifying bacteria (e.g., *Pseudomonas* and *Thiobacillus*) convert nitrates back into Nitrogen gas (N_2) which is them released into the atmosphere.

25.3- The Flow of Energy

Along with the matter, energy also flows between the physical environment and organisms. But, unlike the cycling of matter, the flow of energy is unidirectional.

25.3.1- Trophic Levels

In an ecosystem the organisms are arranged in different feeding groups, known as trophic levels.

1. **Trophic level 1** consists of green plants, algae and photosynthetic bacteria. These are the primary producers of an ecosystem.
2. **Trophic level 2** consists of primary consumers (herbivores) which feed directly on the producers e.g., insects, rabbit.
3. **Trophic level 3** consists of secondary consumers (carnivores) and the parasites of animals which feed on the herbivores e.g., frog, snake.
4. **Trophic level 4** consists of tertiary consumers (animals that eat carnivores) e.g., wolf, hawk.
5. **Decomposers and detritivores** (crabs, vultures, and jackals) make a separate trophic level. They feed on all trophic levels. They consume detritus (dead, decomposing plant and animal material), and fecal matter.

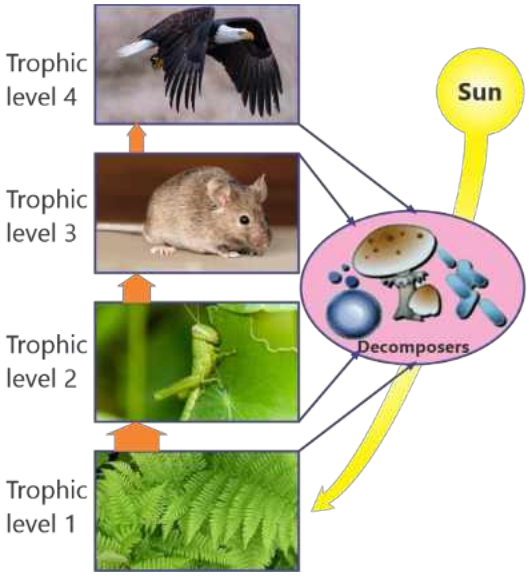


Figure 25.4: Trophic levels

25.3.2- The Loss of Energy between Trophic Levels

There are generally three or four trophic levels in an ecosystem. About 1 to 2% of the solar energy that falls on a first trophic level i.e., producers is converted to the bond-energy of organic matter. During the flow of energy to the next trophic levels, only about 10% is transferred to the next trophic level. Energy is lost at each trophic

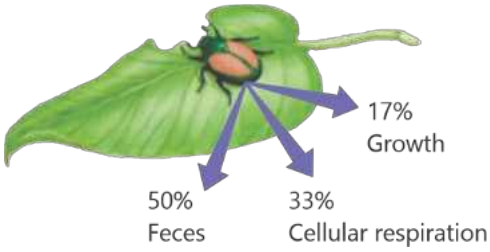


Figure 25.5: Utilization of energy by a heterotroph

level. In this way, very little usable energy remains in the system after it has been used and incorporated into the bodies of organisms at different trophic levels.

For example, if producers fix 10,000 kcal energy by photosynthesis, the primary consumers would get only 1,000 kcal of it. Of these, about 100 kcal are incorporated into the bodies of secondary consumers. If tertiary consumers eat secondary consumers, they gain about 10 kcal of the 10,000 kcal that originally entered the system.

Tidbit

Short food chains (with a smaller number of trophic levels) are more energy efficient than long food chains (with more trophic levels). This is because, energy is lost as heat during energy transfer at each trophic level. So, efficiency is higher when fewer steps are involved in the food chains.

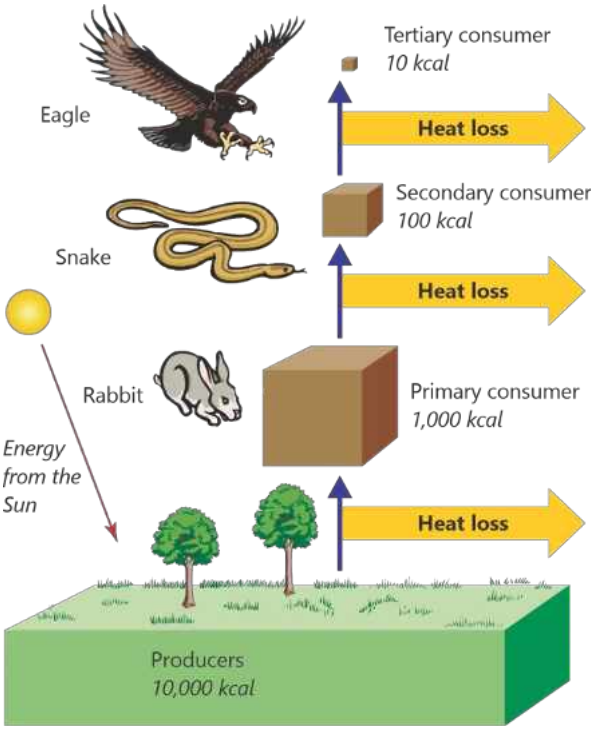


Figure 25.6: Energy flow in trophic levels

25.4- Characteristics of Population

Demography is the statistical study of the characteristics of populations. Population size is the most important features of demography because it is directly related to the ability of a given population to survive.

1- Population Survivorship

Population survivorship is the percentage of members of a group that are alive at a given age. It is often visualized via survivorship curves in survivorship graphs. In such graphs, the Y-axis shows the numbers of survivors, and the X-axis mentions age. Such graphs make three kinds of survivorship curves.

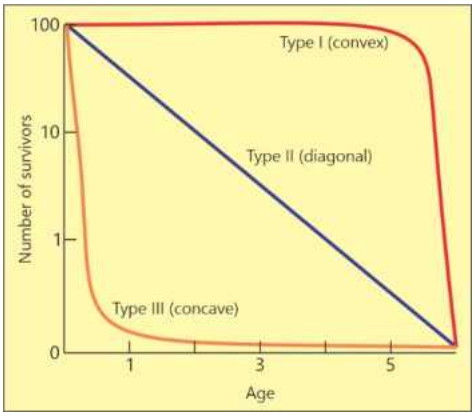


Figure 25.7: Survivorship curves

1. **Type 1 (convex) populations:** In such populations, individuals survive to an old age, then die rapidly. Environmental factors have very less impact on mortality

most individuals live their potential life span. Humans' and mammals' populations make type I survivorship.

1. **Type II (diagonal) populations:** In such populations, individuals have a constant probability of death throughout their lives. The environment has an important influence on death and is no harsher on the young than on the old. Populations of birds, lizards, and rodents exhibit type II survivorship curves.
2. **Type III (concave) populations:** In such populations, individuals experience very high juvenile mortality rates. Those reaching adulthood, however, have a much lower mortality rate. Fishes and many invertebrates display type III survivorship curves.

2- Population Growth

The Increase in the number of individuals within over a specific time period is called population growth. It is an important attribute of populations. There are two population growth models.

Exponential growth model: According to this model, the population grows without limits at its maximal rate. Rather than increasing by adding a constant number of individuals to the population in every generation, the population increases by the same ratio per unit time. Not all populations display exponential growth.

Logistic growth model: In this model, the growing population eventually reaches a limit due to shortage of resources like space, light, water, or nutrients. Such population stabilizes at a certain size, called the carrying capacity of the particular place where it lives. In these situations, growth curves assume a sigmoid, or flattened S, shape.

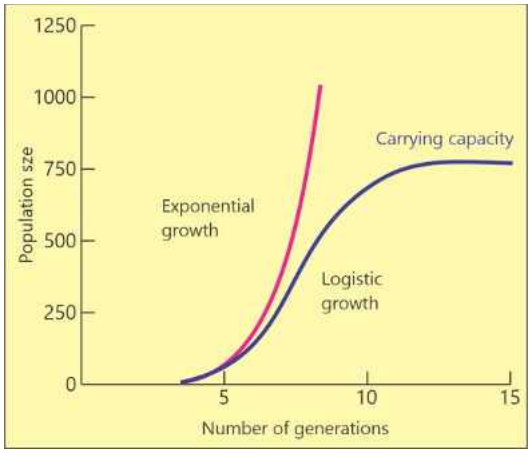


Figure 25.8: Population growth models

3- Population Density

Population density is the number of individuals per unit area or volume. e.g., the number of mulberry trees per square kilometer in Muzaffarabad or the number of *E. coli* bacteria per milliliter in a test tube. In populations with high density, when populations approach their carrying capacity, competition for resources can be severe,

leading to decreased birth rate and increased risk of mortality. High population densities can also result in high rates of pollution.

4- Population Dispersion or Distribution

Another key characteristic of population structure is the way in which individuals of a population are dispersed or distributed within the population.

1- Random spacing: In random spacing, the individuals within populations do not interact strongly with one another. It is not common in nature. For example, it occurs in some plants that have wind-dispersed seeds that germinate wherever they fall in a favorable environment.

2- Uniform spacing: In some populations, individuals are uniformly distributed. For example, some plant species inhibit the growth of nearby individuals, leading to a uniform distance between each plant. Similarly, animals that maintain defined territories, such as nesting penguins, also exhibit uniform dispersion.

3- Clumped spacing: In some populations, individuals clump into groups or clusters due to uneven distribution of resources. Clumped spacing is common in nature because individual animals, plants, and microorganisms tend to prefer resourceful habitats. Clumped spacing is seen in plants that drop their seeds straight to the ground, or animals that live in groups.

5- Population Carrying Capacity

The carrying capacity is the maximum number of individuals of a particular species that a specific environment can support sustainably over a long period. Carrying capacity is determined by "limiting factors" such as the availability of food, water, shelter, and nesting sites, as well as the presence of predators and diseases.

6- Minimal Viable Population Size

The minimal viable population size is the smallest number of individuals required for a population to survive and persist in the ecosystem. If a population falls below this critical size, it faces a high risk of extinction. In a very small population (like the few remaining Himalayan Brown Bears in Pakistan) inbreeding occurs. This leads to genetic weaknesses and a lower immunity against diseases. Moreover, a single natural disaster (e.g., flood or forest fire) may wipe out a population that is below its minimal viable size.

Day of Eight Billion

While it took 12 years for the global population to grow from 7 to 8 billion. In 2022, it reached 8 billion. Now, it will take about 15 years (until 2037) to reach 9 billion. By 2050, the global population will reach nearly 10 billion, with the vast majority still residing in developing regions.

25.4.1- Rapid Growth of Human Populations

Starting in the early 1700s, changes in technology have given humans more control over their food supply, enabled them to develop superior weapons to ward off predators, and led to the development of remedies against many diseases. At the same time, humans have become less vulnerable to climatic uncertainties. These changes allowed humans to expand the carrying capacity of their habitats.

About 16% (1.28 billion) of the world's population lives in developed countries, while the remaining 84% (6.72 billion) reside in developing countries. According to the Pakistan Bureau of Statistics, the last census in 2023 recorded Pakistan's population to be 241.49 million. Pakistan's population growth rate is officially recorded at 2.55% annually.

For Information
According to the UNO, the population of Pakistan has now crossed 250 million. Pakistan maintains its rank as the 5th most populous country in the world.

Effect on Ecosystem

The rapidly growing human population is perhaps the greatest challenge to the future of the biosphere. It is placing severe strains on the global environment. The following are some of these impacts:

- Habitat destruction and changes in land use
- Depletion of natural resources (freshwater, fossil fuels etc.)
- Poverty and inflation
- Increased waste generation and elevated pollution
- Climate change
- Deforestation and loss of biodiversity
- Increased chances of new epidemics and pandemics, starvation and malnutrition

For Information
If you weigh all life on Earth, the human population only makes up about 0.01% of the biomass on Earth. About 82% of the Earth's biomass is made by plants while bacteria make about 13% of the biomass.

For Information
The wealthiest 20% of the world's population consumes 86% of the world's resources and produces 53% of the world's carbon dioxide emissions. Whereas, the poorest 20% of the world consumes only 1.3% resources and emits only 3% of CO_2.

25.5- Greenhouse Effect

Greenhous effect is the natural process where gases present in Earth's atmosphere trap the heat radiated from Earth's surface. The gases are called Greenhouse Gases (GHGs). These gases allow sunlight to pass through while prevent heat from escaping directly back into space. So, they keep the Earth warm enough to sustain life.

There has been a marked increase in the concentrations of GHGs, particularly CO_2. It is mainly due to activities in which fossil fuels are used for running machinery,

driving vehicles, heating homes and generating electricity. The enhanced, unnatural warming of Earth's atmosphere caused by excessive GHGs is the major negative impact of human activities on our environment.

Major Greenhouse Gases

Important greenhouse gases include:

1. **Carbon Dioxide (CO_2):** It is the most significant GHG. It is released through burning fossil fuels (coal, oil, and gas) and deforestation. It contributes about 80% of global warming.

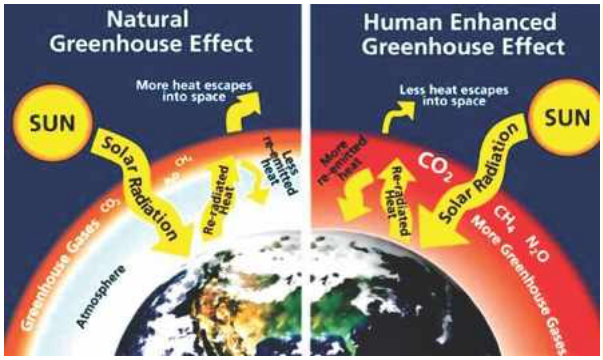


Figure 25.9: Natural and enhanced greenhouse effect (a cause of global warming)

Tidbits

According to the World Meteorological Organization (WMO), before industrialization (1900) the global atmospheric CO_2 concentration was about 280 parts per million (ppm). It has elevated to 428 ppm in 2026 and continues to rise.

The actual mean global temperature has increased about 1°C since 1900, a change known as global warming.

2. **Methane (CH_4):** It is produced during the production of coal and gas, and by livestock (enteric fermentation in cattle) and agricultural practices. It contributes about 11% of global warming.
3. **Nitrous Oxide (N_2O):** It is emitted during agricultural and industrial activities, and during combustion of fossil fuels. It contributes about 6% of global warming.
4. **Fluorinated Gases (Chlorofluorocarbons):** These are synthetic gases emitted from industrial processes (like old refrigerators and air conditioners). These gases contribute about 3% of global warming.

Harmful Effects of Greenhouse Gases

1- **Global Warming:** The intensified greenhouse effect due to emission of excessive GHGs primarily causes global warming. It refers to the long-term increase in the Earth's average surface temperature due to human activities. According to the Intergovernmental Panel on Climate Change (IPCC) Report (2023), global surface temperature has increased by roughly 1.1°C as compared to the pre-industrial era (1850–1900).

Tidbits

For Pakistan, which contributes less than 1% of global GHG emissions but is ranked among the top 10 most vulnerable countries to the devastating effects of climate change.

2- **Heatwaves:** Global warming has turned cities like Jacobabad and Sibi into some of the hottest places on Earth. When atmospheric temperature crosses 50°C, the

human body cannot adjust normal body temperature, leading to fatal heatstroke. Livestock also experiences heat stress. Winters become shorter and warmer allowing pests and mosquitoes to survive longer. Moreover, heat can worsen the draught conditions. The hot, dry weather increases the risks of wild fires.

3- **Floods:** A warmer atmosphere holds more moisture, leading to erratic and intense rainfall. This leads to erratic and "turbo-charged" monsoons and floods. The floods cause significant damage to forests, homes, and agriculture. They also cause long term damage such as soil erosion, nutrient depletion, and contamination of water resources. Floods also lead to rapid pathogen growth and spread of water-borne diseases.

Figure 25.10: The floods in parts of Pakistan in 2022

Tidbits

In June 2025, intense rains in Pakistan caused worst floods. These floods causes over 1,000 deaths, displacement of over 3 million people, destruction of over 200,000 homes, widespread agricultural and infrastructure losses across all provinces and Gilgit Baltistan. This disaster was due to heavy pre-monsoon rains, glacial-lake outburst floods, and cloudbursts.

4- **Glacial Melting:** Elevated greenhouse effect causes melting of glaciers. Pakistan has more glaciers than anywhere outside the polar regions. Rising temperatures cause these glaciers (like Baltoro and Batura) to melt rapidly. This leads to **Glacial Lake Outburst Floods**. In 2025, several such bursts occurred in Gilgit-Baltistan and Chitral, destroying bridges and entire villages. In the long run, once the glaciers vanish, the Indus River—the lifeline of our agriculture—could run dry.

Tidbits

A **Glacial Lake Outburst Flood** (GLOF) is a sudden release of water from a glacial lake due to the breaking of its natural dam (e.g., ice).

The water of GLOF can travel over 100 km from its source, affecting areas far downstream.

A **cloudburst** is a sudden and extremely heavy rainfall in a short period over a small area.

5- **Sea Level Rise and Coastal Erosion:** As polar ice melts and seawater expands due to heat, sea levels rise. For example, the coastal areas in Thatta and Badin are threatened, where seawater intrusion is already turning fertile agricultural land into salty marshes.

6- **Food Insecurity:** High temperatures during the flowering stage of Wheat and Rice can significantly reduce grain yield. Similarly, intense heat evaporates soil moisture, turning fertile lands into deserts (Desertification).

7- **Ecosystem Disruption:** Rapid shifts in temperature and rainfall patterns cause serious threats to biodiversity. Such disruptions force the species for changing their habitats or migrate. It further leads to species threatening and even extinction.

25.6- Important Ecosystems of Pakistan

Pakistan's geography ranges from the highest peaks in the north to the Arabian Sea in the south. It creates a "miniature world" of ecological diversity. The following are the four most significant ecosystems of Pakistan.

25.6.1- Temperate Deciduous Forest

These forests are found in the moist mountain regions where there are distinct seasons, including a cold winter that causes trees to drop their leaves (deciduous).

- **Location:** Northern areas including **Shogran, Neelam Valley, Murree,** and parts of **Kaghan**.
- **Abiotic Profile: Soil:** Rich, grayish-brown soil with high organic matter (humus).
- **Climate:** Moderate temperatures (4°C to 30°C); annual rainfall between **750 mm and 1500 mm**.
- **Key Flora:** *Taxus baccata* (Yew), *Pinus wallichiana* (Blue Pine), *Cedrus deodara* (Himalayan Cedar), and shrubs like *Berberis lyceum* (Sumbal).
- **Key Fauna:** Rhesus monkey (*Macaca mulatta*), Black bear, Leopard cat, wolves and various bird species.
- **Current Status: Degraded.** Many large predators have been wiped out, and the forest is under pressure from the lumber industry and agricultural expansion.

Figure 25.11: A temperate deciduous forest with representative flora and fauna

25.6.2- Tropical Thorn Forest

This is the most widespread terrestrial ecosystem of the plains in Pakistan. It is characterized by xerophytic (drought-resistant) plants with thorns to deter herbivores and reduce water loss.

- **Location:** The Indus Plains, covering much of **Punjab** (Bahawalpur, Khanewal, Mianwali, Dera Ghazi Khan), **Sindh** (southern regions bordering the Indus River), and **Balochistan** (western regions).
- **Abiotic Profile: Climate:** Arid to semi-arid; summer temperatures can reach **50°C**.
- **Water:** Scanty rainfall (200 mm to 750 mm annually).
- **Key Flora:** *Prosopis cineraria* (Jhand/Kandi), *Salvadora oleoides* (Peelu/Wan), and *Capparis aphylla* (Karir) and *Vachellia nilotica* (Kikar).
- **Key Fauna:** Camels, houbara bustard, chinkara, golden jackal, Balochistan gerbil, porcupines, and rattlesnakes.
- **Current Status: Heavily Degraded.** Most of these "natural jungles" have been cleared for human settlements and the massive canal-irrigation agriculture of the Punjab.

Figure 25.12: A tropical thorn forest with representative flora and fauna

25.6.3- Alpine and Sub-Alpine Meadows

These are high-altitude "cold deserts" or grasslands, where the climate is too harsh for large trees to grow.

- **Location:** Mountain tops of Gilgit-Baltistan, Chitral, Swat, Azad Jammu & Kashmir, and the Deosai Plains.
- **Abiotic Profile: Temperature:** Severe cold; mean annual temperature between 1°C and 10°C. **Water:** Precipitation mostly in the form of heavy snow (up to 6 feet).
- **Key Flora:** Stunted Junipers, Birch (*Betula utilis*), willow, and various alpine grasses and wildflowers.
- **Key Fauna:** Snow leopard, Himalayan brown bear, Himalayan Ibex, musk deer and Marmots. Birds like snowcock, monal, golden eagle are also found here.
- **Current Status: Fragile.** These areas are highly sensitive to Global Warming, as melting snow caps alter the delicate balance of the meadow

Figure 25.13: An alpine Meadows with representative flora and fauna

25.6.4- Mangrove Forest (Coastal Ecosystem)

Often called "The Forests of the Sea" these are salt-tolerant forests located at the interface of land and sea.

- **Location:** Sindh (Keti Bandar and Mirpur Sakro mangroves in Indus River Delta) and along the Balochistan coast (Miani Hor, Kalmat Khor, Jiwani & Gawatar Bay).
- **Abiotic Profile: Water:** Brackish (a mix of fresh river water and salty seawater). **Soil:** Anaerobic (oxygen-poor) mudflats; trees have "breathing roots").
- **Key Flora:** Avicennia marina (Timmar) makes up 95% of the forest.
- **Key Fauna:** Crocodiles, shrimps, mudskippers, crabs, lobsters, turtle, snakes, dolphins and birds like flamingo, crane, pelican.
- **Current Status: Threatened** by a reduction in freshwater flow from the Indus and pollution from Karachi, though recent restoration projects have helped save the area.

Figure 25.14: A mangrove forest with representative flora and fauna

EXERCISE

SECTION 1: MULTIPLE CHOICE QUESTIONS

35. If you study the interaction between a herd of Markhors, the grass they eat, and the snow leopards that hunt them, which level of ecological organization are you focusing on?

    (a) Species (b) Population (c) Community (d) Biosphere

36. Nitrogen-fixers are present in the group/s;

(a) Bacteria and fungi (b) Bacteria only
(c) Fungi only (d) Bacteria, fungi and viruses

37. In an ecosystem, the energy flow is;
(a) Always unidirectional (b) Always bidirectional
(c) Non-directional (d) Cyclic

38. Absorption and re-emission of heat radiation by the atmosphere is called;
(a) Global warming (c) Ozone depletion
(c) Acid rain (d) Greenhouse effect

39. A population's carrying capacity.
(a) Can be accurately calculated
(b) Generally, remains constant over time
(c) May change as environmental conditions change
(d) Can never be exceeded

40. During energy flow in ecosystems, how much of the energy at a trophic level enters the next level?
(a) 100% (b) 50% (c) 25% (d) 10%

41. Which group of bacteria is involved in nitrification?
(a) *Azotobacter* (b) *Nitrosomonas*
(c) *Clostridium* (d) *Rhizobium*

42. Which step of nitrogen cycle is accomplished by *Pseudomonas*?
(a) Ammonification (b) Nitrification
(c) Nitrogen fixation (d) De-nitrification

43. How do nitrogen-fixing bacteria contribute to the nitrogen cycle?
(a) Return nitrogen (N_2) to the atmosphere
`(b) Change ammonium to nitrate
(c) Change N_2 to ammonium
(b) Absorb nitrate from the soil

44. According to 2025-2026 climate data, the "Enhanced Greenhouse Effect" is considered as climate injustice for Pakistan because;
(a) Pakistan produces the majority of global greenhouse gases.
(b) Greenhouse Effect only occurs in South Asia.
(c) Pakistan emits less than 1% of global GHGs but suffers extreme warming and floods.
(d) Industrialized nations have successfully stopped all carbon emissions.

SECTION 2: SHORT QUESTIONS

1. What are the primary reservoirs of the chemicals of biogeochemical cycles?
2. Write notes on de-nitrification and ammonification.
3. Briefly describe the role of bacteria in the nitrogen cycle. Enlist their names and roles.

4. Write a brief note on the flow of energy in trophic levels.
5. Write brief notes on population density and population growth.
6. Investigate the effects of human population growth on the environment.
7. How does the increased greenhouses gases in atmosphere result in global warming?
8. What are the long-term effects of global warming?
9. Describe the unique adaptations of flora in the Mangrove ecosystem.
10. Distinguish between the Abiotic profiles of a Temperate Deciduous Forest and a Tropical Thorn Forest.
11. Enlist the key fauna and flora of temperate deciduous forests, tropical rain forest, and mangrove forest.
12. Differentiate between:
    - Biome and biosphere
    - Biogeochemcial cycle and energy flow
    - Population growth and population density
    - Convex population and concave population
    - Nitrification and nitrogen-fixation
    - Nitrification and denitrification

SECTION 3: LONG QUESTIONS

31. What is a biogeochemical cycle? Describe water cycle in detail.
32. Describe nitrogen cycle in detail.
33. Explain the flow of energy in successive trophic levels.
34. Explain the problems related to the rapid growth of human populations and the effects of that growth on ecosystem.
35. Describe the causes and impacts of the increasing concentration of greenhouse gases in the atmosphere.
36. Provide a comprehensive account of the Tropical Thorn Forest of Pakistan. Give its location, key flora and fauna, and its current ecological state.
37. Write a detailed note on the Temperate Deciduous Forest in Pakistan.

INQUISITIVE QUESTIONS

18. Justify the fact that humans are responsible for climate change.
19. Investigate the effects of human population growth on ecosystem.
20. Justify why science education has become necessary for everyone to understand the steps man has to take to save the biosphere.
21. Investigate the careers related to the study of environmental resources.
22. **Activity:** Search internet and collect the pictures of the key fauna and flora of different ecosystems in Pakistan.

Biology Paper Pattern and Pairing Scheme (12th Class) (New 2026-27)

Total marks = 85
The paper is divided into two main sections:
Objective and Subjective.
Time Allowed: 3 Hours

1. Objective Part (MCQs)
Total Marks: 17
Total MCQs: 17
Time Allowed: 20 Minutes

Q.1 MCQs

Marks breakdown

Chapter No:	Chapter Name	No. of MCQs
CHAPTER# 13	(Thermoregulation and osmoregulation)	1 MCQ
CHAPTER# 14	(Human Urinary System)	1 MCQ
CHAPTER# 15	(Human Nervous System)	2 MCQs
CHAPTER# 16	(Human Endocrine System)	1 MCQ
CHAPTER# 17	(Human Reproductive Systems)	1 MCQ
CHAPTER# 18	(Inheritance)	2 MCQs
CHAPTER# 19	(Chromosomes and DNA)	2 MCQs
CHAPTER# 20	(Biotechnology)	2 MCQs
CHAPTER# 21	(Immunity)	1 MCQ
CHAPTER# 22	(Biostatistics)	1 MCQ
CHAPTER# 23	(Pharmacology)	1 MCQ
CHAPTER# 24	(Evolution)	1 MCQ
CHAPTER# 25	(Ecology)	1 MCQ

2. Subjective Part
Total Marks: 68
Time Allowed: 2 hours and 40 Minutes

Marks breakdown

Section I: Short Questions (44 Marks)

Q. 2:(Short Questions): Attempt any 8 out of 12 (Chapters: **13,14, 19, 20**) **2X8=16**

CHAPTER # 13	Short Questions: 3
CHAPTER # 14	Short Questions: 3
CHAPTER # 19	Short Questions: 3
CHAPTER # 20	Short Questions: 3

Q. 3: (Short Questions): Attempt any 8 out of 12 (Chapters: **15, 16, 17, 18, 24**) **2X8=16**

CHAPTER # 15	Short Questions: 2
CHAPTER # 16	Short Questions: 2
CHAPTER # 17	Short Questions: 2
CHAPTER # 18	Short Questions: 4
CHAPTER # 24	Short Questions: 2

Q. 4: (Short Questions): Attempt any 6 out of 9 (Chapters: **21, 22, 23, 25**) **2X6=12**

CHAPTER # 21	Short Questions: 2
CHAPTER # 22	Short Questions: 2
CHAPTER # 23	Short Questions: 3
CHAPTER # 25	Short Questions: 2

Section II: Long Questions (24 Marks)

Attempt any 3 questions out of 5.
Each question consists of two parts: (a) 4 marks and (b) 4 marks.

Q.5: Chapter 13 & 25
Q.6: Chapter 14 & 24
Q.7: Chapter 15 & 21
Q.8: Chapter 16 & 20
Q.9: Chapter 17 & 19

MODEL PAPER OF BIOLOGY FOR CLASS-12

Objective Type

Biology **Time Allowed: 20 min.**

Paper: I (Objective Type) **Maximum Marks: 17**

Q.1. Four possible answers A, B, C and D to each question are given. The choice which you think is correct; fill that circle in front of that question with marker or ink pen in the answer book. Cutting or filling two or more circles will result in zero mark in that question. **[17 × 1 = 17]**

	QUESTIONS	(A)	(B)	(C)	(D)
1	Positive feedback loops are less common because they:	Are slow	Destabilize the system	Use no effectors	Never stop
2	Efferent arterioles of juxtamedullary nephrons give rise to:	Peritubular venules	Afferent arterioles	Vasa recta	Interlobular arteries
3	Long-term drug use reduces dopamine receptors on:	Presynaptic neuron	Postsynaptic neuron	Axon terminal	Synaptic vesicles
4	Which disorder is autoimmune in nature?	Alzheimer's disease	Huntington's disease	Multiple sclerosis	Parkinson's disease
5	Glycoprotein hormones are unable to pass through the membrane because they are:	Lipid-soluble	Non-polar	Polar	Steroid
6	LH stimulates:	Sertoli cells	Germ cells	Leydig cells	Epididymis
7	Crossing over may alter expected ratios by:	preventing segregation	creating linkage breaks	increasing dominance	reducing fertilization
8.	A woman is a carrier for haemophilia. What percentage of her sons may inherit the disease?	25%	50%	75%	100%
9	Okazaki fragments are joined by:	Helicase	Ligase	Primase	Gyrase
10	GC hairpin structure mainly functions to:	Start transcription	Stop RNA polymerase	Repair DNA	Translate protein
11	Which vector is specially designed for large DNA fragments?	BAC	tRNA	mRNA	Operon
12	Most reliable method to confirm gene presence is:	Antibiotic test	Blue-white screening	DNA hybridization	Microscopy
13	Why are natural TILs often ineffective against malignant melanoma?	They cannot divide	Tumour suppresses immune response	They lack nuclei	They produce antibodies only
14	Which group in an experiment receives the treatment?	Control group	Dependent group	Experimental group	Independent group

15	The first step in drugs discovery is;	Lead optimization	Preclinical Testing	Target identification	Hit identification
16	Changes in gene frequency due to migration of individuals are called;	Genetic drift	Mutation	Gene flow	Genetic recombination
17	Which group of bacteria is involved in nitrification?	Azotobacter	Nitrosomonas	Clostridium	Rhizobium

MODEL PAPER OF BIOLOGY FOR CLASS-12

Subjective Type

Biology

Paper: II (Essay Type)

Time Allowed: 2.40 Hrs.
Maximum Marks: 68

(SECTION – I) **(22 x 2 = 44)**

Q.2. Write short answers to any eight (8) questions. **8X2=16**

i. How do control centers help maintain homeostasis?
ii. What are the physiological adaptations to reduce water loss in terrestrial animals?
iii. Analyze why humans excrete uric acid even though they are not uricotelic.
iv. Differentiate the two types of nephrons.
v. What is glomerular filtration?
vi. How does ADH relate to Diabetes Insipidus?
vii. Why does DNA have a strong affinity for histone proteins?
viii. Evaluate Griffith's hypothesis about the role of the polysaccharide coat in virulence.
ix. What is a pulse-chase experiment?
x. What is meant by a palindromic sequence in restriction sites?
xi. How the vector turns into recombinant vector?
xii. Differentiate between blue colonies and white colonies in blue-white screening.

Q.3. Write short answers to any eight (8) questions. **8X2=16**

i. Define neuroglia. Write its function.
ii. Name any two excitatory neurotransmitters and state where they act.
iii. What are the functions of Luteinizing Hormone (LH)?
iv. What is myxoedema? What are its symptoms?
v. Name the accessory glands of male reproductive system and their functions.
vi. Why do primary oocytes remain inactive for many years in females?
vii. Justify why O-negative individuals are called universal donors and AB-positive individuals are called universal recipients.
viii. Write brief note on Y-linked inheritance in humans.
ix. How can pleiotropy be considered a limitation to the law of independent assortment?
x. What is Se gene? Where it is located?
xi. Differentiate between Natural selection and neutral selection.
xii. State Hardy-Weinberg theorem.

Q.4. Write short answers to any six (6) questions. **6X2=12**

i. How does respiratory tract provide first line defence?
ii. How does the membrane-attack complex (MAC) destroy microbes?
iii. Why is biostatistics important in medical research?
iv. Describe the difference between descriptive and inferential statistics?
v. Differentiate between antiviral and antiretrovirals drugs.
vi. Write a mode of Tetracycline action?
vii. Write any two advantages of monoclonal antibodies.
viii. Write notes on ammonification.
ix. Investigate the effects of human population growth on the environment.

(SECTION – II)

Note: Attempt any three questions. **(3X8=24)**

Q.5. a) Classify the animals on the bases of the ability to thermoregulate. 4

b) Explain the flow of energy in successive trophic levels. 4

Q.6. a) How are Kidney stones treated? 4

b) Describe the evidences of evolution that come from comparative anatomy. 4

Q.7. a) Explain the events and propagation of action potential. 4

b) How do inflammatory response act as non-specific response to infections? 4

Q.8. a) Describe negative feedback mechanism. 4

b) Explain the steps of polymerase chain reaction (PCR). 4

Q.9. a) Describe the events of a menstrual cycle. 4

b) Explain the experimental work conducted by Griffith that demonstrated that DNA is the hereditary material. 4