

M.Sc. Final Year
Zoology, Paper - V

MICROBIAL ECOLOGY AND
BIOLOGY OF PARASITISM



मध्यप्रदेश भोज (मुक्त) विश्वविद्यालय – भोपाल
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| Unit - II Microbial Ecology <ol style="list-style-type: none">1. Role of microbes in Biogeochemical and Nutrient Cycling processes. Ex. Carbon, Nitrogen, Sulphur and Phosphorus Cycle.2. Microbes in Metal Cycling.3. Aquatic Micro - organisms : fresh water microbiota, Sea water microbiota.4. Role of Micro - organisms in Water purity and water purity test.5. Sewage waste water treatment. 6. Methods used in environmental studies. | Unit-2: Microbial Ecology II (Pages 61-114) |
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INTRODUCTION

Microbial ecology explores the diversity, distribution, and abundance of microorganisms, their specific interactions, and the effect that they have on ecosystems. Although not traditionally thought of as a central discipline within ecology, microbial ecology is of critical importance because microorganisms represent the vast majority of the genetic and metabolic diversity on the planet and drive most of the critical ecosystem processes which recycle matter and energy. Microorganisms have evolved to occupy almost every conceivable ecological niche and energy-generating mechanism. During this process microorganisms engage in a wide range of ecological interactions with each other and with higher forms of life.

Microbial ecology is a branch of microbiology which emphasize on understanding the interactions of microbes with their environment, rather than their behavior under artificial laboratory conditions. Because microbes are ubiquitous, microbial ecologists study a broad diversity of habitats that range from aquatic to terrestrial to plant- or animal-associated. Many different microbial ecosystems can be defined, based upon the association with particles, the presence of environmental gradients, and the continuous availability of water. Microbial ecology not only deals with the microorganism's population ecology and physiological ecology, but also their broad versatility and quantitative importance in the biosphere as biogeochemical catalysts and capacity for rapid physiological and evolutionary responses.

Parasitism is a close relationship between species, where one organism, the parasite, lives on or inside another organism, the host, causing it some harm, and is adapted structurally to this way of life. Like predation, parasitism is a type of consumer-resource interaction, but unlike predators, parasites, with the exception of parasitoids, are typically much smaller than their hosts, do not kill them, and often live in or on their hosts for an extended period. Parasites of animals are highly specialized, and reproduce at a faster rate than their hosts. Classic examples include interactions between vertebrate hosts and tapeworms, flukes, the malaria-causing *Plasmodium* species, and fleas. Parasitism is significant biological process in which one of the partners (the parasite) is physiologically dependent on the other (host). In medicine, parasitism encompasses diseases produced by protozoa, helminthes, and arthropods that are housed or live temporarily or permanently in humans.

This book is divided into four units that attempt to give the students an in depth knowledge of metabolic diversity of microbes, microbes living in extreme environments, ecological role of microorganisms, plant and animal microbe symbiosis role of microbes in biogeochemical and nutrient and metal cycling processes, aquatic micro – organisms, role of micro - organisms in water purity sewage waste water treatment, biology of parasitism, host parasite relationship, protozoan parasites pathogenic to man , Platyhelminthes parasites, Nematelminthes parasites, ectoparasites, parasitic adaptations in parasites, and evolution of parasitism. The

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book follows the Self-Instructional Mode or SIM format wherein each unit begins with an 'Introduction' to the topic followed by an outline of the 'Objectives'. The detailed content is then presented in a simple and structured manner interspersed with Answers to 'Check Your Progress' questions. A list of 'Key Terms', a 'Summary' and a set of 'Self-Assessment Questions and Exercises' is also provided at the end of each unit for effective recapitulation.

UNIT 1 MICROBIAL ECOLOGY I

Structure

- 1.0 Introduction
- 1.1 Objectives
- 1.2 An Introduction to Metabolic Diversity of Microbes
- 1.3 Microbes Living In Extreme Environments and Their Potential for Use in Biotechnology
- 1.4 Ecological Role of Microorganisms in General
- 1.5 Animal Microbe Symbiosis
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- 1.6 Micro-Organisms in Formation of Different Soils
- 1.7 Answers to 'Check Your Progress'
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1.0 INTRODUCTION

Microbial ecology (or environmental microbiology) is the ecology of microorganisms, their relationship with one another and with their environment. It concerns the three major domains of life, Eukaryota, Archaea, and Bacteria as well as viruses.

Microbial life is amazingly diverse and microorganisms quite literally cover the planet. Microbes live in all parts of the biosphere where there is liquid water, including soil, hot springs, the ocean floor, acid lakes, deserts, geysers, rocks, and even the mammalian gut. Most types of microbes remain unknown. It is estimated that we know less than 1% of the microbial species on Earth. Yet microbes surround us everywhere: air, water, soil. An average gram of soil contains one billion (1,000,000,000) microbes representing probably several thousand species. Microbes are vital to every ecosystem on Earth and are particularly important in zones where light cannot approach (that is, where photosynthesis cannot be the basic means to collect energy). Microorganisms participate in a host of fundamental ecological processes including production, decomposition, and fixation. They can also have additional indirect effects on the ecosystem through symbiotic relationships with other organisms. In addition, microbial processes can be co-opted for biodegradation or bioremediation of domestic, agricultural, and industrial wastes, making the study of microbial ecology particularly important for biotechnological and environmental applications.

In this unit you will study about metabolic diversity of microbes, the microbes living in extreme environments and their potential for use in biotechnology, the ecological role of microorganisms, plant microbe symbiosis, animal microbe symbiosis and the role of micro-organisms in formation of different soils.

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1.1 OBJECTIVES

After going through this unit you will be able to:

- Understand metabolic diversity of microbes
- Analyze the microbes living in extreme environments and their potential for use in biotechnology.
- Evaluate the ecological role of microorganisms
- Explain plant microbe symbiosis
- Understand animal microbe symbiosis.
- Analyze the role of micro-organisms in formation of different soils

1.2 AN INTRODUCTION TO METABOLIC DIVERSITY OF MICROBES

Microorganisms are extremely widespread, however, within the microbial world there is also a remarkable number of different metabolic pathways. This happens as microbes consumes as well as produce a variety of compounds. In the last decade, scientists have been able to sequence the whole genomes of several microorganisms, giving them access to the sequences of all the genes present. This offers a sight into the metabolic potential of a microbe as knowing the genes present can suggest which enzymes the microbe can make and use for its metabolic activities. The micro-organism, as a group, conduct all the same types of basic metabolism as higher eukaryotic organism, however, in addition, there are several types of energy-generating metabolism among the prokaryotes that are non-existent in eukaryotic cells or organisms. The diversity of prokaryotes is expressed by their great variation in modes of energy generation and metabolism, and this feature allows prokaryotes to flourish in all habitats suitable for life on earth. Four broad categories of metabolic diversity include:

- (A) The main energy-gathering strategy used by the microbes or energy generating metabolism
- (B) Strategies for obtaining carbon
- (C) Making primary metabolites and essential enzymes for growth
- (D) Biosynthesis of secondary metabolites

(A) Energy Generating Metabolism

There are three sources of energy in nature:

- Organic chemicals (Organic chemicals are those containing carbon–carbon bonds)
- Inorganic chemicals (Inorganic chemicals are those without carbon–carbon bonds)
- Light Chemo-organotrophy is the type of metabolism where energy comes from organic chemicals, i.e., compounds containing carbon. Chemolit-

hotrophy refers to the type of metabolism where energy comes from inorganic chemicals (chemical compounds lacking carbon). Phototrophy involves turning light energy into metabolic energy in a process known as photosynthesis, and it comes in two main forms:

- **Oxygenic Photosynthesis**- This kind of photosynthesis generates oxygen and is used by the cyanobacteria (a type of bacteria) and algae (a eukaryote), as well as all living plants.
- **Anoxygenic Photosynthesis**- This kind of photosynthesis does not make oxygen and is used by the purple and green bacteria (types of bacteria that live in anaerobic aquatic environments).

(B) Strategies for Obtaining Carbon

All the living cells requires a lot of carbon, which is part of all proteins, nucleic acids, as well as cellular structures. Organisms that utilize organic carbon are referred to as heterotrophs; chemorganotrophs fall into this category. Organisms that utilizes carbon dioxide (CO₂) for their carbon needs are known as autotrophs; most of the chemolithotrophs as well as phototrophs are also autotrophs, which makes them primary producers in nature as they make organic carbon out of inorganic carbon dioxide that is then available for themselves, chemoorganotrophs, and eventually all higher life forms.

A few organisms can switch between heterotrophy mode of nutrition when organic carbon is available and autotrophic mode of nutrition when food sources run out; these organisms are referred to as mixotrophs.

(C) Making Primary Metabolites/Enzymes

Few compounds in nature are not degraded by microorganisms. The variety of compounds produced by microbes is great and not completely known. Their metabolic processes are essential for environmental nutrient cycling, and they are the primary producers that support all other life on earth. Microbes are experts at degrading a variety of compounds ranging from the simplest to the most complex. Microbes are the only organism capable of degrading *resistant plant material* (fiber) made from *cellulose* (building blocks used by plants to make their tough cell walls) and *lignin* (building blocks used by plants for rigid structure, as in wood and straw). The microbes in the *rumen* (part of a cow's or related animal's stomach) of herbivores as well as the guts of termites are responsible for digesting these tough plant fibers. Fungi and bacteria are the masters of producing special enzymes to degrade complex food sources (hydrolytic enzymes) including all forms of plant and animal tissues, some plastics, and even metals. Primary metabolites are microbial products which are produced continuously during the exponential phase of growth and are involved in primary metabolic processes like respiration as well as photosynthesis. They include intermediates and end-products of anabolic metabolism, which are used by the cell as building blocks for essential macromolecules (For instance: - amino acids, nucleotides) or are converted to coenzymes (For instance: - vitamins). Industrially, the most significant primary metabolites are amino acids, nucleotides, vitamins, solvents and organic acids. Many of these metabolites are manufactured by microbial fermentation and isolation instead of chemical synthesis because the fermentations are economically

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competitive and produce biologically useful isomeric forms. A few characteristic features of primary metabolites are as follows:

- Those chemical compounds which are directly involved in the growth, development as well as reproduction of living organisms are known as primary metabolites.
- Those primary metabolites which are present in most cells throughout the body are termed as central metabolites.
- Primary metabolites are generally produced in large quantities and can easily be extracted from the plants.
- Primary metabolites are a part of the basic molecular structure of an organism.
- Primary metabolites are highly useful in metabolic process of organisms as some act as a substrate for these processes, while others act as catalysts.
- Primary metabolites are directly involved in the process of growth, development as well as reproduction.
- Primary metabolites are not active in the defense mechanism.
- Primary metabolites are synthesized during the trophophase stage of an organism.
- Examples of primary metabolites include Alcohol, amino acids, nucleotides, antioxidants, organic acids, vitamins and polyols.

(D) Biosynthesis of Secondary Metabolites

Secondary metabolites refer to the organic compounds which are not directly involved in the process of growth, development as well as reproduction of the organism. Secondary metabolites are generally produced by fungi, bacteria or plants and have an important ecological function. Secondary metabolites are driven by pathways in which primary metabolites are involved. Therefore, secondary metabolites are considered as the end products of pathways of primary metabolites. Environmental factors do influence the production of secondary metabolites though the regulation of the formation of secondary metabolites is more complex and differs from that of primary metabolites. A few examples of secondary metabolites include: Pigments, alkaloids, drugs, essential oils, antibiotics, ergot alkaloids, nucleosides, quinolines, peptides, phenazines, naphthalenes, terpenoids, lectins, polymeric substances and lectins. The characteristic features of secondary metabolites are as follows:

- Secondary metabolites refers to the organic compounds synthesized by bacteria, fungi or plants which are not directly involved in the normal growth, development as well as reproduction of the organism.
- Secondary metabolites are produced in minute quantities when compared to primary metabolites
- Secondary metabolites are unique in different organism. Secondary metabolites are species-specific and thus are different in different organisms.
- They are not part of the basic molecular structure of an organism.
- The metabolic activities of the organism are not affected by the absence of secondary metabolites

- Examples of secondary metabolites are: - pigments, alkaloids, drugs, essential oils, antibiotics, ergot alkaloids, nucleosides, quinolines, peptides, phenazines, naphthalenes, terpenoids, lectins, polymeric substances and lectins.
- Secondary metabolites are formed during stationary phase of an organism and is known as idiophase.
- Secondary metabolites are involved in defense mechanism
- Secondary metabolites play a role in ecological functions such as serving as antibiotics and producing pigments.
- Many of these products are bioactive compounds useful in interacting with other organisms.
- Secondary metabolites are active against foreign invaders and might be involved as a defense mechanism.
- Microbes make molecules that are useful in communicating with other microorganisms, insects, and plants.

Here are some other ways that the knowledge of microbial metabolism has been useful in the advancements of science:

- **Microbial enzymes are used in molecular biology research**

Enzymes like Taq DNA polymerase (used for reproducing sequences of DNA) and restriction enzymes (used to manipulate pieces of DNA in a cut-and-paste fashion) have become invaluable research tools. These enzymes are used for carrying out process like polymerase chain reaction.

- **Microbes are used to express animal proteins or enzymes like insulin.**

Microbes are involved in the mass production of hormones molecules like Insulin which is used to treat disease like diabetes.

- **Microbial systems are used as part of microscopic machines in synthetic biology**

Microbes are used widely to generate new molecules of potential medical and therapeutic importance. This methodology combines engineering with genetics.

- **Industrial processes have taken advantage of the diversity of microbes in the food, pulp and paper, mining, and pharmaceutical industries.**

As few microorganisms are tolerant of extreme conditions, the enzymes they produce are useful in industrial settings where conditions can be harsh.

Check Your Progress

1. What is chemo-organotrophy?
2. Define Chemolithotrophy.
3. What are mixotrophs?
4. Write some examples of secondary metabolites?

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1.3 MICROBES LIVING IN EXTREME ENVIRONMENTS AND THEIR POTENTIAL FOR USE IN BIOTECHNOLOGY

An extreme environment is a habitat which is characterized by harsh environmental conditions i.e., conditions above or below the optimal range necessary for the growth, development as well as reproduction of the organism. Extreme environments is characterized by several unfavourable conditions like high or low temperature, high or low pressure, and acidic or basic pH. An environment is considered to be harsh when certain conditions or aspects of the environment must be considered very hard for different forms of life to survive. For instance: the polar region, deserts, volcanic regions, deep ocean trenches, outer space, and every other planet of the Solar System except the Earth are considered to be harsh environment for the survival of different forms of life. Areas that are highly alkaline, acidic, extremely hot or cold, high salt concentration, without water or oxygen are considered to be extreme.

On the basis of extreme physiochemical conditions, extreme environments are classified into the following groups: -

(a) Extreme Temperature Environment

Extreme cold and hot are two types of extreme environments on the basis of temperature.

- Environments having a temperature less than 5°C are considered as extremely cold environments. Such extreme cold environments exist in deep ocean niches, at the peaks of high mountains, or the Polar Regions.
- Environments having a temperature higher than 45°C are considered as extremely hot environments. Such extreme hot environments are influenced by geothermal activity as geysers and fumaroles of continental volcanic areas or deep-sea vents.

(b) Extreme pH Environment

On the basis of pH extreme environments can also be classified as acidic or alkaline:

- Those natural habitats having a pH below than 5 are considered as extreme acidic environments
- Those natural habitats having a pH above 9 are considered as extreme alkaline environments

(c) Extreme Ionic Strength

- Natural habitats having an ionic concentration higher than of seawater (greater than 3.5%) are considered as Hypersaline environments.

d) Extreme Pressure

- Those habitats which are under extreme hydrostatic or litho pressure, such as aquatic habitats at depths of 2,000 m or more or deep-subsurface ecosystems are considered as extreme pressure environments.

(e) High-Radiation Environments

- High radiation environments are those habitats that receives high radiation doses, like ultraviolet or gamma radiation. Habitats like deserts and the top of high mountains are considered as high radiation environments.

(f) Xeric Environments

Xeric environments are arid habitats having limited water supply. Cold and hot deserts are few instances of these extreme environments. Example of xeric environments are Sahara Desert.

Extremophiles are living organisms that have the ability to survive and thrive well in extreme environments. This is possible due to different physiological and molecular adaptations in extremophiles. Extremophiles can thrive well in extreme habitat like ice, salt solutions, acidic or alkaline conditions. They are capable of growing in toxic waste, organic solvents, heavy metals, and other such habitats where life is not possible. Most of the extremophiles are prokaryotic organisms, with few eukaryotes as exception. These extremophiles are characterized by the environmental conditions in which they can survive and thrive optimally. Extremophiles can be divided into two categories namely:

- (a) Extremophilic Organisms-** These organisms require one or more than one extreme environmental condition to survive and flourish.
- (b) Extremotolerant Organisms-** These organisms can survive and flourish at neutral conditions, however, they can tolerate extreme conditions of one or more physical parameters.

Extremophiles include members of all three domains of life; bacteria, archaea, and eukarya. Most of the extremophiles are prokaryotes, i.e., organism lacking a definite nucleus such as archaea. However, few eukaryotes like protists (e.g., algae, fungi, and protozoa) and multicellular organisms are also extremophiles. Extremophiles can be categorized on the basis of the conditions in which they grow as follows:-

(a) Psychrophile Organisms

- Psychrophiles are the organism adapted to cold conditions. These microorganisms have been able to colonize permanently cold environments like deep sea to mountain and Polar Regions. The optimum temperature required for growth of psychrophile is around 15°C.
- Environments like ocean waters, permafrost, glaciers, Antarctic rocks, snowfields, and polar ice caps are suitable for the growth of such microorganisms.
- Prokaryotic psychrophiles exhibit different requirement necessary for growth and development like some species are aerobic whereas others are anaerobic or facultative.
- A few examples of psychrophile microorganism include species of Psychrobacter members of Halomonas species, psychrophilic species of Pseudomonas and Alteromonas spp, Hyphomonas spp, Sphingomonas

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spp, *Chryseobacterium greenlandensis*, *Desulfotalea psychrophila*, *Psychrobacter arcticus*, etc.

(b) Thermophile Organism

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- Thermophiles (literally heat lovers) are organisms that grow at temperatures above those (25–40°C) that sustain most life forms. Typically, a thermophile shows maximum growth rates at temperatures above 45°C.
- There are many examples of the environment with extreme temperatures. Environments with high temperatures include both terrestrial and submarine environments.
- Different habitats throughout the world are suitable for the growth of thermophiles. These can range from surface soil to compost piles.
- Thermophiles are generally found in areas like thermal vents, hot springs, and boiling steam vents.
- Thermophiles are further divided into different groups; facultative thermophiles and obligate thermophiles.
- Facultative thermophiles can thrive at both high and moderate temperatures whereas obligate thermophiles require high temperature for growth.
- Like in the case of psychrophiles, thermophiles also have different physiological and molecular adaptations that enable the organisms to survive at temperatures that would normally denature proteins, cell membranes, and genetic material.
- Some of the common examples of thermophiles include *Methanosarcina thermophila*, *Methanobacterium wolfei*, *Methanobacterium thermoautotrophicum*, *Archaeoglobus profundus*, *Alicyclobacillus acidoterrestris*, *A. acidocidarius*, etc.

(c) Hyperthermophile Organisms

- Hyperthermophiles refers to the organisms that can survive and grow at extremely high temperatures above 80°C.
- Hypothermophiles are a type of thermophiles that can tolerate even higher temperatures when compared to other thermophiles.
- Even though, the optimum temperature for growth of hyperthermophiles is 80°C, however, they can survive at temperatures higher than 100°C.
- Apart from this, most of the hyperthermophiles are capable of enduring other extreme conditions like higher pH as well as higher pressure.
- Most of the hyperthermophilic organisms are found in hot springs and boiling steam vents where even moderate thermophiles cannot survive or thrive.
- Most of the hyperthermophiles belong to the archaea group with very few species being bacterial species.
- For instance: *Thermoproteus uzoniensis*, *Staphylothermus marinus*, *Pyrodictium abyssi*, *Pyrococcus furiosus*, *Hypothermus butylicus*, *Pryococcus woesei*, *Pyrodictium brockii*, *Pyrodictium occultum*, etc. are hyperthermophiles.

(d) Acidophile Organisms

- Acidophiles are the organisms that can withstand and thrive at highly acidic conditions having a pH around 2.0.
- Acidophilic microorganisms survive well in extremely low pH natural or artificial environments like acidic lakes, hydrothermal systems, acid sulfate soils, sulfidic regoliths, ores, etc.
- Highly acidic environments are established by the oxidation of the metal as well as other sulfidic minerals that are populated by a range of acidophilic and acid-tolerant prokaryotic and eukaryotic life forms.
- Heterotrophic, acidophilic bacteria, frequently living in close association with chemolithotrophic primary producers, have also been isolated from extremely acidic environments.
- The most broadly studied acidophiles are prokaryotic organism that oxidize reduced iron as well as sulfur.
- They can catalyze the oxidative dissolution of metal sulfide minerals such as pyrite (FeS_2), thereby strictly acidifying the environment (having a pH less than 3) in which they thrive.
- There are several natural acidic environments that include volcanic areas, hydrothermal sources, deep-sea vents, metal mining areas, as well as the stomachs of animals.
- All three domains of life i.e., archaea, bacteria, and eubacteria have few acidophilic organisms but archaea denote the major acidophilic group of organisms.
- Physiologically, the acidophiles are very diverse: aerobic and facultative anaerobic, chemolithotrophs, and different types of heterotrophic prokaryotes, photoautotrophic eukaryotes, predatory protozoa, and others.
- For instance: *Lactobacillus*, *Thiobacillus sulfolobus*, *Bacillus acidocaldarius*, *Thermoplasma acidophilus*, *Picrophilus*, *Ferroplasma acidiphilum*, *Acidithiobacillus*, *Leptospirillum*, *Acidobacterium* spp., *Sulfobacillus*, etc. are acidophilic organism.

(e) Alkaliphile Organisms

- Alkaliphiles refers to a group of extremophiles that can survive and in environments having extremely high pH value. Even though, the optimal pH for growth and survival of these organisms is around 10, yet they can tolerate pH ranging from 9-13.
- Alkaliphiles are of two types namely obligate alkaliphiles and facultative alkaliphiles. Obligate alkaliphiles can grow only in environments with pH higher than 9, however, facultative alkaliphiles can live both at neutral pH as well as alkaline conditions.
- Majority of the organisms growing under high alkaline conditions are described to be prokaryotes. Prokaryotes comprises of a heterogeneous collection of eubacteria along with archaeobacteria.

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- Apparently, alkaliphiles can be easily isolated from ‘normal’ environments like garden soil as transient alkaline conditions have been observed in such environments due to biological activity.
- These organisms are believed to be the earliest life forms on Earth which originated billions of years ago in deep-ocean alkaline hydrothermal vent systems.
- Alkaliphiles survive and flourish well in many geographical locations across the planet, both in natural and manmade alkaline environments like alkaline soda lakes, soda deserts, and saline soda soils.
- Other natural habitats in which alkaliphiles flourish include the alkaline serpentine lakes, oceanic bodies, ikaite tufa columns, and alkaline hydrothermal vents.
- For instance: *Bacillus alkaliphilus*, *Bacillus pasteurri*, *Bacillus halodurans*, *Halobacterium*, *Clostridium paradoxum*, *Halomonas pantelleriensis*, *Alkaliphilus hydrothermalis*, etc. are alkaliphiles.

(f) Xerophile Organisms

- Xerophile refers to a group of organisms that are capable of surviving and flourishing well in conditions having low availability of water or low water activity.
- Usually, xerophilic organisms are capable of growing at a_w values lower than xerotolerant organisms (a_w below 0.8). a_w values refer to water activity levels.
- In conditions where very little amount of water is available, organisms must take up as well as maintain sufficient water against extreme concentration gradients to support cellular processes.
- Xerophiles are of dissimilar types belonging to different groups of living beings.
- Xerophilic fungi represent a large group of xerophilic organisms.
- Eukaryotic organisms such as plants are capable of surviving at low water condition and are referred to as xerophytes are also categorized as xerophiles.
- Xerophiles are very closely related to halophiles as halophilic environments tend to have low water activity.
- Although, water is essential for numerous biomolecular processes in living beings, xerophiles have intricate means to survive in conditions with low water activity.
- **For instance:** *Aspergillus penicillioides*, *Cereus jamacaru*, *Deinococcus radiodurans*, *Aphanothece halophytica*, *Anabaena*, *Bradyrhizobium japonicum*, *Saccharomyces bailli*, etc. are few examples of xerophiles.

(g) Halophiles

- Halophiles refers to a group of extremophiles that require high salt concentrations for their growth as well as survival.

- Halophiles are of two types namely obligate halophiles that require NaCl concentration of 3% or more and halotolerant that survive at both average salt concentrations and higher.
- Halophilic microorganisms comprise of the natural microbial communities of hypersaline ecosystems, which are broadly distributed across the world.
- Halophiles are characterized by low nutritional requirements as well as high resistance to hypersaline environment.
- Halophiles are categorized based on their salt requirement as: Slightly halophile, moderately halophile and extreme halophile. Slightly halophile requires low salt (1-3%); moderate halophile requires moderate amount of salt (3-15%), and extreme halophile requires high concentration of salt (15-30%).
- However, salt requirement of a halophile depends on a lot of other factors such as temperature, pH, as well as growth medium.
- Halophiles are physiologically diverse
- Halophiles can be aerobic as well as anaerobic, heterotrophic, phototrophic, and chemoautotrophic.
- The habitat of halophiles ranges from hypersaline soils, springs, salt lakes, sabkhas to marine sediments.
- These organisms are widely present in all three domains of life, i.e., Archaea, Bacteria, and Eukaryota.
- Halophilic bacteria are more abundant in specific phylogenetic subgroups, most of which belong to Halomonadaceae, a family of Proteobacteria.

Halophilic organisms in terms of their salt requirement are as follows:-

1. **Slightly Halophilic:** *Erwinia*, *Bacillus hunanensis*, *Halomonas zhaodongensis*, *Alkalibacterium thalassium*, etc.
2. **Moderately Halophilic:** *Spiribacter salinus*, *Halobacillus sediminis*, *Halobacillus salicampi*, *Marinobacter piscensis*, *Idiomarina aquatica*, etc.
3. **Extreme Halophile:** *Halococcus salifodinae*, *Halobacterium salinarum*, *Limimonas halophilia*, *Lentibacillus kimchii*, *Sporohalobacter salinus*, etc.

(h) Osmophiles

- Osmophiles refers to a group of organisms that are capable to survive in environments with high osmotic pressures like high sugar concentration.
- Hence, they like halophiles as well as xerophiles are capable of flourishing in environments with low water activity.
- They have been observed majorly in food with high sucrose content as well as environments with high osmolarity.
- The most common example of osmophiles are Fungi.
- However, organisms of the group Archea and Bacteria are also important osmophiles.

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- Most of the osmophiles maintain a higher osmotic pressure in the cytoplasm as compared to surrounding environment, leading to an outward-directed pressure, turgor, whose maintenance is essential for cell division as well as growth.
- Any variations in environmental osmolarity can initiate the flux of water across the cytoplasmic membrane.
- Thus, osmophilic organisms develop different mechanisms to overcome the osmotic imbalance.
- For instance: *Zygosaccharomyces*, *Torula*, *Schizosaccharomyces octosporus*, etc.

(i) Piezophiles/ Barophiles

- Barophiles refers to a group of organisms that are capable of growing and flourishing optimally at pressures greater than atmospheric pressure.
- The term piezophile is often used as a replacement to barophile as piezo means pressure in Greek.
- These bacteria have been isolated from various deep-sea environments throughout the world.
- These organisms exhibit rapid growth at low temperatures and high pressures.
- Majority of the barophilic organisms tend to be psychrophilic and thus cannot be cultured at a temperature above 20°C.
- Further, several barophiles tend to be obligate barophiles with few archaea acting as moderately barophilic.
- Few instances:- *Shewanella benthica*, *Moritella yayanosii*, *Shewanella violacea*, *Photobacterium profundum*, *Moritella japonica*, *Sporosarcina* spp, etc.

(j) Endolith/Hypolith

- Endolith refers to group of organisms that can survive in several inhospitable environments like inside rocks, animal shells, coral reefs, as well as sand particles in the soil.
- Endoliths occupy habitats beneath as well as between porous and translucent rocks and minerals.
- Rock porosity provides interstitial spaces for microbial colonization as well as translucence enables photosynthesis to take place.
- Cold deserts also harbor endolithic microorganisms.
- Endolithic microbial communities have been extensively studied in the Antarctic region, known for its extreme climatic conditions, such as low humidity and precipitation.
- In terrestrial systems, these microenvironments typically provide protection from intense solar radiation as well as desiccation.
- In a marine system, endolithic microbes exploit the rocky seafloors, however, it also dwell into limestone and mineralized skeleton of a wide variety of marine animals.

- Endothilic organisms are also subjected to osmotic stress due to the high salt concentrations.
- For instance: *Leptolyngbya*, *Helicobacter recurvirostre*, *Gloeocapsa sanguine*, *Acaryochloris*, *Chroococciopsis*, *Anabaena*, *Spirirestis rafaensis*, etc.

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(k) Hypoliths

- Hypoliths refers to a group of organisms or communities of organisms that live on the underside of rocks or at the rock–soil interface.
- They are photosynthetic microorganisms that occur in hot as well as arid climates, generally at the interface of the rocks and the soil.
- These microorganisms are protected from the harsh radiations of the sun and the wind.
- The moisture trapped by rocks helps such microorganism to flourish.
- A variety of minerals like quartz are found in soil and rocks which supports different forms of life.
- The most common habitats for hypoliths include the desert lands and polar regions
- For instance: *Nostoc*, *Bryum*, *Hennediella*, *Stichococcus mirabilis*, *Ichthyosporea*, etc.

(l) Metallotolerant

- Metallotolerant refers to a group of microorganisms that are capable of tolerating as well as detoxifying high levels of dissolved heavy metals.
- These microorganisms utilize metals as structural components of biomolecules, or as cofactors in several reversible oxidation/reduction reactions, or in electron transfer chains during energy conservation.
- Still, metals can become toxic if their intracellular concentrations get too high.
- Majority of the metallotolerant microorganisms tend to be acidophilic in nature as the physiological activities of metallotolerant organism enable tolerance against high metal concentrations.
- Metallotolerant microbes can be aerobic and facultative aerobic chemoheterotrophic microorganisms.
- Polluted soils as well as polluted waters containing untreated industrial and urban wastes (with a high concentration of metals) are important habitats of metallotolerant microorganisms.
- For instance: *Bacillus subtilis*, *Bacillus megaterium*, *Acidithiobacillus ferrooxidans*, *Acidithiobacillus caldus*, *Corynebacterium diphtheriae*, *Acidiphilium rubrum*, *Acidiphilium multivorum*, etc.

(j) Radiophiles

- Radiophiles refers to a group of extremophiles that are capable of surviving the extreme forms of radiations like ionizing radiation (gamma rays) as well as UV radiation.

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- Only a few studies are conducted on radiophiles till date as they need to be isolated from extreme environments like outer space of other planets.
- Radiophiles are less diverse. All radiophiles belongs to the archaea and bacteria families.
- Radiophiles can either be radiation tolerant or radiation-resistant.
- Radiation resistant microorganisms can survive harmful radiation for a long period of time as compared to radiation tolerant organism.
- For instance: *Deinococcus radiodurans*, *Brevundimonas*, *Rhodococcus*, *Halomonas*, *Herbaspirillum*, *Hymenobacter*, *Rhodobacter*, etc.

Application and Potential Use of Extremophiles in Biotechnology

- Enzymes are the natural products that complete chemical reactions in an ultra-efficient as well as non-polluting way. In the context of food and environmental crisis, demanding the development of a bio-inspired economy, the new enzymes present in the genomes of populations of extremophilic microorganisms (and called extremozymes) are of great attention
- In fact, the ability of the extremozymes to perform chemical reactions under extreme conditions and sometimes the uniqueness of the chemical reactions they perform make them very interesting for multiple applications.
- Extremophilic enzymes serve as a model systems to study as well to conduct research studies on various aspects of enzymes such as enzyme evolution, enzyme stability, activity, mechanism, protein structure, function, and biocatalyst under extreme conditions.
- Thermophiles have yielded stable α -amylase for starch hydrolysis.
- Thermophiles have yielded oxylonases for paper bleaching
- Thermophiles have yielded proteases for brewing as well as for detergent purposes.
- Alkaline active proteases, amylases, cellulases, mannanases, lipases, etc. are used in the formulation of heavy-duty laundry as well as dishwashing detergents.
- Such detergents are highly efficient in removing stains and allow effective low-temperature (30–40°C) washing.
- A few species of acidophilic microorganisms can be used effectively to reduce mine water pollution.
- Further, acidophilic organisms can be used to recover metals from acidic wastewater via selective bio-mineralization.
- Enzymes like Taq polymerase obtained from *Thermus aquaticus* is one of the most helpful biotechnological products derived from extremophiles. It is used in polymerase chain reaction as it reduces the need for adding extra polymerase during the reaction.
- Cellulose for various extremophilic organisms has been used for the treatment of juices, color brightening in detergents, and treating cellulose-containing biomass and crops to improve their digestibility and nutritional quality.

- Halophiles have been developed as a possible source of carotene, compatible solutes, glycerols, as well as surfactants for pharmaceutical use.
- A few extremophilic microorganisms may also include a large reservoir of novel therapeutic agents—for instance, iron-binding antifungal compound, pyochelin isolated from halophilic species of *Pseudomonas*.
- A thermostable glucokinase from the thermophilic species, *Bacillus stercorophilus*, can be used as a glucose sensor for quick glucose assay.
- Alkaline active enzymes have been recognised in textile and fibre processing in processes like cotton scoring and blast fibre degumming.
- Alkaliphiles as well as their enzymes have been tried in numerous synthesis reactions with peptide synthesis being the most significant one.
- Information related to the microbial composition as well as biogeochemical cycling of extreme ecosystems also helps in clearly understanding the global change, threats, and opportunities for living beings.
- The enzymes derived from extremophiles can also be used effectively in several bioremediation processes such as removing metallic waste from sewages and industries.
- Enzymes derived from barophilic organism can be used for the production and sterilization of items at varied pressure conditions.
- Biotechnologists use extremozymes for the production of biofuels, biomaterials or pharmaceutical molecules.
- Halophilic enzymes are capable of operating in saline environments, in organic solvents and in a wide range of pH.
- These enzymes have been used effectively in industrial and commercial processes like food processing, in the paper industry as well as in the textile industry.
- Thermozyms and barozymes from thermophilic and/or barophilic organisms are hyperstable enzymes that can be used for food applications under conditions that eliminate the risk of bacterial contamination.
- They can be used under physico-chemical conditions corresponding to multiple processes used by the textile, leather, cosmetic or pharmaceutical industries.
- Due to their originality, abundance and the many interactions and symbioses that govern the dynamics of bacterial and Archaean communities in extreme environments, these microbes represent a largely unexplored genetic resource.
- Hence, the search for new biocatalysts as well as antibiotics based on the microbial biodiversity of extreme environments is a quickly growing discipline needing the development of dedicated enzymatic and structural screening and characterization platforms.

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- Check Your Progress**
5. What do you understand by extreme environment?
 6. Define xeric environments.
 7. What are Extremophiles?
 8. What is the optimum temperature required for growth of psychrophile?
 9. Which group of organisms are capable to survive in environments with high osmotic pressures?

1.4 ECOLOGICAL ROLE OF MICROORGANISMS IN GENERAL

Microorganism is a living body of a small size and exist everywhere in our surrounding. All creatures having size of several microns or smaller can be said as microorganisms. These organisms can be unicellular or multicellular, so that including the group of microorganisms are bacteria, viruses, protozoa, archaea, algae, and fungi (Dwidjoseputro, 2003). Thus, microorganisms are small, living creatures which are invisible to our naked eye. Hence, they are also known as microscopic organisms. Among all microorganisms, some are harmful and disease-causing pathogens, while others include useful microbes, which are more beneficial and harmless to humans.

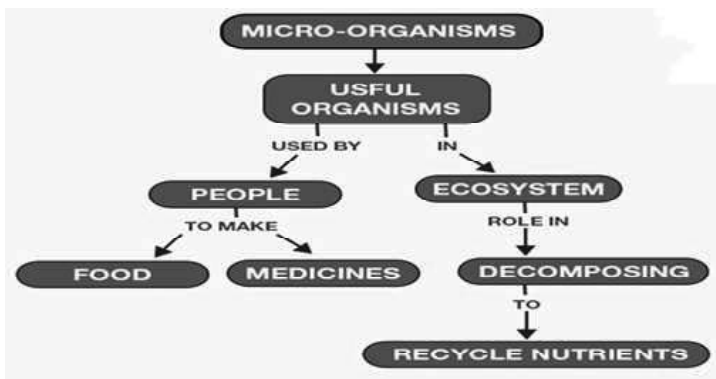


Fig.1.1 Role of Microorganisms in Various Fields

Microorganisms are ubiquitous, i.e., they exist everywhere on or in Earth, be it water, soil or air. The study of microbes that exist in their natural environment is called as microbial ecology. Ecology is part of biology related to the study of relationships of organisms or groups of organisms with their environment.

In this section, we shall discuss about the ecological role of microorganisms.

(1) Role of Microbes in Aquatic Environment

Aquatic microorganisms include members of the bacteria, protozoa as well as fungi. These organisms differ radically, and share only their small size; most are not visible without a microscope, though colonies of some can be seen with the naked eye. Microorganisms are present in large quantities everywhere and can survive

extreme physical and chemical conditions. Many microorganisms play foundational roles in aquatic ecosystems, capturing the sun's energy through photosynthesis and, through their role in decomposition, releasing nutrients stored in organic tissue. For instance: Protists are microscopic organisms present in abundance in the ecosystem. Protists can be both plant like and animal like. Plant like protists serves as producers and manufacture food by the process of photosynthesis whereas animal like protists (paramecia, Amoeba) feeds upon bacteria and other small protists, so they form part of the food-chain. A third category of protists, i.e., fungus like protists often serve as decomposers in the ecosystem. Fungi depends upon the bodies of their living hosts or depends upon remains of once living organisms. Fungi, also serves important role in the ecosystem as decomposers. Bacteria acts as decomposers in the ecosystem, i.e., they have the ability to break down remains of dead and decaying organisms for the release of nutrients in the ecosystem. They can also act as producers, for ex in deep-sea vents, chemosynthetic bacterial fills the role of producers in food chain. Further, bacteria also serve as food material for other organism like protists. All surface waters such as ponds, lakes, rivers and oceans differ in their physical, chemical and biological characters. Depending on the nutrient status of water body, the microbial load varies with higher numbers encountering in eutrophic waters. Ground waters or subterranean waters generally have very low microbial load because of filtration effect of soil layers. The natural waters contain a huge variety of microorganisms. These include,

- Natural flora: Microorganisms natural to the water body and
- Transient flora: Microorganisms entering the water body from outside environment like from soil, air and through pollutants.

Microorganisms present in natural aquatic environment play huge role in different ecological phenomenon like nutrient recycling, primary producers and decomposers of organic matter. All these microorganisms present in an aquatic system can be observed as surface flora of inhabiting organisms. These not only include spoilage organisms but also human pathogenic microorganisms especially in sewage contaminated waters. The role of microorganism present in aquatic bodies are as follows

(a) Ecological Role

In freshwater system like lakes, ponds and streams, plants as well as certain microbes perform photosynthesis and harvest the energy of the sun. Microbial photo-synthesizers include algae and cyanobacteria which are considered as primary producers in the aquatic system. Other organisms referred to as consumers, feed on these organisms and form a link in the food chain. Decomposers are third in the category, and represent special kind of organisms in the food chain. They consume dead bodies of plants, animals, and other microbes and convert detritus (dead and decaying matter) and organic matter into simple nutrients, such as nitrate, phosphate, and sulfate. Decomposers are significant for major biogeochemical cycles by which nutrients are exchanged between living and non-living parts of the ecosystem. Without microbial decomposers, it would be impossible for minerals and nutrients to reach flora and fauna of aquatic system. These minerals and nutrients are critical for the growth and survival of plant as well as animal.

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NOTES**(b) Microbes as Producer of the Aquatic System**

Microbes that act as producers are microbes that can make food (synthesis of organic material) itself through the process of photosynthesis. The kind of microbes that act as producer commonly from the group of algae that float in the water, usually known as phytoplankton. Phytoplankton and periphyton are the chief primary producers in lentic water system. Phytoplankton are seen drifting in the water column of the pelagic zone. However, several phytoplanktons have a higher density and sink to the benthic region. Periphytons remains attached to the substrate covering almost the entire benthic surfaces. Phytoplanktons as well as periphyton serves as important food source and chief oxygen producers in the lentic system. Phytoplankters are small microscopic, unicellular and photosynthetic organisms which freely float in water bodies. Phytoplanktons composed of both eukaryotic as well as prokaryotic species which colonizes upper euphotic part of the water column ranging from freshwater to ocean conditions. They also exhibit amazing adaptation to remain in floating condition. The range of the Phytoplankton cells can vary in size from about 1 μm to 1 mm. Like terrestrial plants, these tiny primary producers need sunlight, nutrients as well as carbon dioxide for their growth & multiplication. Phytoplankters cells contains chlorophyll which helps them to synthesize food in the presence of sunlight. Therefore, they help in maintaining not only the carbon dioxide level but also help to alleviate global warming to some extent. Physical process like wind and current play substantial part in their distribution particularly in estuarine and marine conditions. Phytoplankton act as primary link in energy pathway to higher trophic level via several food chains.

(c) Microbes as Decomposer of Aquatic System

Decomposer microbes can degrade specific compounds and have a role in biogeochemical cycles in water. Bacteria are present in all regions of aquatic water system. Free-living bacterial forms are associated with decomposing organic material, or are present on the surfaces of rocks as well as plants, suspended in the water column, and in the sediments of the benthic and profundal zones. A few other bacterial forms are associated with guts of lentic animals as parasites or in commensal relationships. Bacteria play an essential function in system metabolism via nutrient recycling. Majority of bacteria in lentic water system depends upon dead and decaying organic matter to obtain their energy. These bacteria are then eaten up by protozoa which in turn are consumed by zooplankton and then further up the trophic levels.

(d) Role of Microbes in Food Chain and Food Web Operated in the Aquatic System

Ecosystem is defined as the interaction between the biotic and abiotic components in any given area. These interactions are essential for maintaining the flow of energy that cycles from the abiotic environment and travels through living organisms via food-web. This energy flow is ultimately transferred back to the abiotic environment when living organisms die and the cycle starts all over again. All the biotic and abiotic components of the ecosystem are closely interlinked with each other and thus there is a continuous flow of energy from abiotic to biotic components through different trophic levels arranged in the form of pyramid. The energy and matter produced by autotrophic organisms are distributed to other organisms in an ecosystem through pathways known as food chains and food webs.

In a marine ecosystem, phytoplankton provide food to tiny shrimp called krill which then is feasted upon by blue whale.

The trophic level defines an organism's position in the food chain. For instance: A fish feeding on an insect, which itself has just consumed algae, is at a higher trophic level than the insect. On the contrary, food web depicts a network of linked food chains, since usually organisms consume, and are consumed by more than one type of organism. Food webs connect autotrophs, which form the very base of the web, to the herbivores (primary consumers) and then to various carnivores (secondary consumers). In a river system, as well as in the majority of other aquatic and terrestrial systems, the energy at the base of a food web comes from the sun and it is fixed by autotrophic organisms via the process of photosynthesis. Energy from the sun is transferred through the ecosystem by passing through various trophic levels. The energy pyramid has a large base and becomes smaller at the top. At the base or the first level are the producers. Here, the energy and the number of organisms (biomass) are very high. At the second level are the various consumers. For each level or feeding stage, the energy and number of organisms decrease so the pyramid becomes smaller at the top. Energy derived from terrestrial plants enters in to the aquatic system in the form of plant parts, like leaves or twigs, or in the form of dissolved organic matter. This plant material is utilized as a source of energy by microorganisms like fungi and bacteria, and also by invertebrates. Microscopic algae are often eaten while alive, while larger aquatic plants mainly enter food chains after they have died. Further, in the event of oil spill, the number of bacteria and fungi that break down detritus and provide nutrients for autotrophs will change leading to further changes at all levels of the food web. Oil spills leads to the formation of a thick layer of oil on the surface of water. This thick oil layer can stop marine plants from receiving enough light for carrying out the process of photosynthesis as well as it can disturb the free flow of oxygen in the water. This severely affects the floral and faunal biodiversity.

(e) Role in Nutrient Cycling

The matter is recycled via Earth's ecosystems, and it may move from one ecosystem to another as it does when nutrients are washed away into a river. The same atoms are used over and over again, assembled into different chemical forms and incorporated into the bodies of different organisms.

For instance: Primary producers like plants, algae, cyanobacteria occupies the first trophic level. These autotrophs take in carbon dioxide from the atmosphere as well as other nutrients, like nitrogen and phosphorous, from the water to build complex molecules that finally make up their cells. When a heterotrophic animal eats the primary producer, it utilizes its molecules to obtain energy and as building material for its own cells, often rearranging atoms and molecules into new forms. Such heterotrophic animals which eat up the autotrophs directly are known as herbivores or primary consumers. These herbivores or primary consumers represent the second trophic level. These first level animals are eaten up by next level consumers known as carnivores or secondary consumers. These secondary consumers or carnivores represent the third trophic level. The top carnivores are called tertiary consumers, and they represent the fourth trophic level. When aquatic plants and animals carry out cellular respiration they break down complex molecules

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as fuel, releasing the carbon dioxide into the atmosphere. Similarly, when they excrete waste or die, their chemical compounds are used for energy and building material by bacteria and fungi. These decomposers release simple molecules back into the water and atmosphere, where they can be taken up by primary producers.

Energy, unlike matter, cannot be recycled in ecosystems. Energy flow through an ecosystem is a one-way street, usually, from light to heat. Energy usually enters ecosystems as sunlight and is captured in form of a chemical compound by photosynthesizers like plants and algae. The energy is then passed through the ecosystem, changing forms as organisms metabolize, produce waste, eat one another, and eventually, die and decompose. Each time energy changes forms, some of it is converted to heat. Heat still counts as energy, and thus no energy has been destroyed, but it generally can't be used further as an energy source by living organisms. Ultimately, the energy that entered the ecosystem as sunlight is dissipated as heat and radiated back into space. This one-way flow of energy through ecosystems means that every ecosystem needs a constant supply of energy, usually from the sun, in order to function.

An ecological pyramid is a graphical representation of the relationship between the different living organisms at different trophic levels. It was given by G.Evlyen Hutchinson and Raymond Lindeman.

Pyramid of energy is always upright. Pyramid of energy represents the total amount of energy present at each trophic level and it also accounts for the energy lost at each step while moving up to another trophic level. Hence the pyramid is always upward, with a large energy base at the bottom. For instance: - an ecosystem receives approximately 1000 calories of light energy in a given day from sun. Out of this, only 1% i.e., around 100 calories are stored as energy-rich materials by algae and phytoplankton (autotrophic organism). Now, next a primary consumer eats the plant containing 100 calories of food energy. The primary consumer will utilize a part of this energy to carry on its physiological and metabolic activities and hence stores only 10 calories as food energy. Next, a secondary consumer that eats up the primary consumer gets an even smaller amount of energy. Thus, usable energy keeps on decreasing from one trophic level to other, i.e., from primary producers to primary consumers to secondary consumers or top consumers. Therefore, the energy pyramid will always be upright.

(f) Excessive Growth of Microorganism Leads to Oxygen Deficient Aquatic Bodies

Eutrophication refers to the process in which a water body becomes overly enriched with nutrients, leading to excessive growth of growth of algae and plankton. The excessive growth (or bloom) of algae and plankton in a water body are indicators of this process. Eutrophication is considered to be a serious environmental matter as it often leads to the deterioration of water quality and the depletion of Dissolved Oxygen (DO) in aquatic bodies. Eutrophic waters can eventually become "dead zones" that are no longer capable of supporting aquatic life (both plants and animals). Over a period of time, anthropogenic activities like use of excessive fertilizers in agricultural field, disposal of nutrient rich waste into water bodies etc. has increased the nutrient content of freshwater bodies ultimately leading to the deterioration of water quality and making it unfit for human consumption.

(g) Dissolved Oxygen Levels in the Aquatic System

DO is the abbreviation used for Dissolved Oxygen. Dissolved oxygen characterizes the concentration of oxygen in an aqueous solution and refers to the free oxygen dissolved in water. The unit of dissolved oxygen is mg/L, expressed in milligrams of oxygen per litre of water. The amount of dissolved oxygen in the water is an indicator of the self-purification ability of the water body. High dissolved oxygen is conducive to the degradation of various pollutants in the water body, so that the water body can be purified faster; on the contrary, when the dissolved oxygen is low, the pollutants in the water body degrade slowly. The concentration of oxygen in inland waters depends upon factors within the water. Oxygen is released into the water bodies by biological production or in simple terms by the process of photosynthesis carried out by autotrophs as well as by simple aeration methods i.e. passage of oxygen through the air-water interface, however, dissolved oxygen is consumed by the process of biological decay and advanced metabolism oxygen consumption. The mutual ebb and flow of these two effects makes the dissolved oxygen content in water show temporal and spatial changes.

NOTES**(h) Biological Oxygen Demand**

The amount of organic matter present in the water can be assessed by supplying oxygen to the water sample under observation. The organic matter present in water will start consuming the oxygen and undergoes oxidation. Thus, Biological Oxygen Demand (BOD) or Biochemical Oxygen Demand (BOD) is referred as the amount of oxygen needed or demanded by aerobic microorganisms to break down the organic matter present in a certain sample of water at a specific temperature over a given period of time. BOD of water sample is evaluated by measuring the oxygen consumed by the bacteria from the decomposition of organic matter over a period of five days at a specific temperature of 20 degree Celsius. A lot of factors influence the BOD of water like temperature, pH of the water sample, inorganic substance present in the water, kind of aerobic microorganisms present in water, amount and type of organic matter present in the water sample etc. BOD values indicate the polluting potential of water. A BOD value of less than 1mg/L indicates pure drinking water, BOD value between 2-8mg/L indicates moderately polluted water whereas a BOD value greater than 8mg/L indicates severely polluted water unfit for consumption. BOD finds its prime significance in sewage treatment plants. It indicates the respiration rate in sludge, soil, garbage etc.

(i) Clarity of the Water

Turbidity refers to the measure of relative clarity of a liquid. It is an optical characteristic of water and is a measurement of the amount of light that is scattered by material present in the water when a light is passed through the water sample. Turbidity is directly proportional to light scattered i.e. higher the intensity of scattered light, higher is the turbidity of water. The material that increases the turbidity of water includes slit, tiny organic matter, inorganic material, clay, plankton, dissolved matter as well as other microscopic organism. Higher turbidity makes the water cloudy or opaque. In fact, turbidity of the water is an indicator of potential pollution present in the water bodies. Excessive turbidity, cloudiness or opaqueness in drinking water is not only unappealing, but also unhealthy. High turbidity can provide habitat for pathogens which may lead to outbreak of water borne diseases.

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As mentioned above, turbidity of water directly influences the penetration of light. It limits the penetration of light in the aquatic bodies. Productivity of the fresh water system depends upon the penetration of the light. For instance:- Large shallow lakes tend to be more productive when compared to lakes with deep water as more light reaches the lake bed. In larger rivers, the canopy is open which allows the algal production.

(2) Microbial Degradation of Petroleum

Petroleum and its products are the hydrocarbons. Petroleum is a rich source of organic matter and gets oxidised easily on coming in contact with air or moisture. There are few microorganisms which can cleave the hydrocarbons into simpler molecules. For instance: Fungi and bacteria are the major agents which can decompose oil and oil products. Besides fungi and bacteria, cyanobacteria, yeast and algae have shown to oxidise hydrocarbons. The simplest hydrocarbon pollutant is methane. It is degraded by a specialized group of bacteria known as methanotrophic bacteria. Fats/Oils are insoluble in water and keeps on floating over the surface of water to form slicks or oil films. Microorganisms capable of oxidising hydrocarbons develop rapidly in such films. Oil is present both in anoxic (absence of O_2) as well as oxic (presence of O_2) environment as is evidenced by the presence of natural oil deposits. Numerous *Pseudomonas*, different cyanobacteria, various corynebacteria as well as mycobacteria are capable of degrading petroleum products. Initially, the non-volatile components are oxidised by bacteria and in the later process, certain fractions of branched-chain and polycyclic hydrocarbons are degraded slowly. The microbial production of hydrocarbon occurs in colonial alga, *Botryococcus braunii*, by secretion of long-chain hydrocarbons (C_{30} to C_{36}) that have the consistency of oil. An increasing interest has been shown in using this type of microbe as renewable sources of production of petroleum.

(3) Waste Treatment

Microbes (fungi and bacteria) traditionally serves as a decomposer. In the absence of microbial decomposer, planet earth will be filled by the carcass in large quantities. Microbial decomposer is used for the processing of garbage / waste. All the waste material can be divided into organic and inorganic waste. Inorganic or Non-organic garbage will be recycled, while organic waste would have continued the process of composting. Microbes plays an essential role in treating the organic waste as well as help in waste water treatment. For instance: - Biological treatment of wastewater is done to obtain water fit for human consumption. Biological treatment involves the extensive use of micro- bacteria that decompose the waste material or organic impurities and improve the quality of water to be used for households. Three common methods of biological treatment are as follows: -

- (a) **Aerobic Process:** As the name suggests, oxygen is required for this purpose. During the aerobic process, oxygen consuming bacteria decompose and devour the organic impurities of wastewater and convert it to CO_2 for reproduction and growth.
- (b) **Anaerobic Process:** This process occurs in the absence of oxygen. In the Anaerobic or fermentation process, waste or sludge present in the wastewater is fermented at a specified temperature.

- (c) **Composting:** Composting is a kind of Aerobic process, in which sludge or biosolid is mixed with carbon sources in the presence of oxygen to remove all the impurities.

(4) Role of Microbes in Terrestrial Environment

Microbes play an essential role in providing nutrients for plants. A few essential roles of terrestrial microorganism are as follows:

a. Generate Oxygen in Atmosphere

- Almost all of the oxygen produced in earth today occurs by bacteria in prehistoric period.
- Most of the oxygen producers are cyanobacteria (blue green algae) in ocean.
- Oxygen is released by the plants during photosynthesis. Humans and other animals inhale the oxygen exhale carbon dioxide which is again taken up by the plants. They utilise this carbon dioxide in photosynthesis to produce oxygen, and the cycle continues.

b. Recycle Nutrition Stored in Organic Matters to Inorganic Form

- Sulphur cycle moves via the rocks, water bodies and living systems. Sulphur is released into the atmosphere by the weathering of rocks and is converted into sulphates. These sulphates are then taken up by the microorganisms as well as plants. Sulphates are converted into organic forms by microorganisms as well as plants. Organic sulphur is consumed by animals via food. When the animals die and decompose, sulphur is returned to the soil which is again obtained by the plants and microbes, and the cycle continues.
- Phosphorus moves via the hydrosphere, lithosphere and biosphere. Phosphorus is extracted by the weathering of rocks. Due to the phenomenon of rains and erosion, phosphorus is washed away in the soil and water bodies. Plants and animals obtain this phosphorus from the soil via water for their growth and survival. Microorganisms also require phosphorus for their growth. When the plants and animals die they decompose, and the stored phosphorus is returned to the soil and water bodies which is again consumed by plants and animals and the cycle continues.
- Thus, microbial decomposition of organic matters releases the minerals such as N, P, K etc. and these minerals are bound up and made available for producers (green plants) to use. Without this recycling primary productivity of ecosystem would stop.
- In soil, fungi are the most important decomposers of plant materials and are followed by bacteria.

c. Fix atmospheric Nitrogen into Useable Form and Increasing the Fertility of Soil

Nitrogen Fixation

Nitrogen is required by plants to produce proteins, DNA, RNA, ATP, NAD⁺, NADP⁺, and Chlorophyll, etc. Nitrogen fixation refers to the conversion of nitrogen into nitrate, a form that can be used by plants. Nitrogen fixation can be carried out by volcanic action, lightning, industrial activity and by some bacteria. Nitrogen-

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fixing bacteria are present freely in the soil or live in the root nodules of leguminous plants like peasbeans, clover. The nitrogen fixing bacteria use the plant as a carbohydrate source of energy, protection and shelter (anaerobic conditions too) and the plant uses the nitrate produced by the bacterium (e.g., of mutualism). This is the first step of the nitrogen cycle and is characterized by the conversion of atmospheric N_2 into ammonia (NH_3). Bacteria like Azotobacter and Rhizobium have a major role in this process. They are harboured in the roots of the leguminous plants and help convert inert nitrogen to ammonia. Further, Organic remains of plants and animals are broken down in the soil by some bacteria to release ammonia into the soil. This dead and waste matter is used by these microorganisms as food and they release ammonia into the soil. This is another process by which ammonia can be generated

Nitrification

In this step, the ammonia is converted to nitrites and then to nitrates by nitrifying bacteria. Some of the nitrate formed in the soil is absorbed and assimilated by the plants. This occurs in two-steps. The first step is in which NH_3/NH_4^+ is converted to NO_3^- (nitrates). The bacteria Nitrosomonas and Nitrococcus present in the soil convert NH_3 to NO_2^- , and another bacterium, Nitrobacter converts NO_2^- to NO_3^- . These bacteria gain energy through these conversions. The nitrates are taken up by plants to form DNA, Protein etc. The process is known as assimilation.

Denitrification

The conversion of nitrates to nitrogen gas. It is carried out by denitrifying bacteria in the soil. These bacteria are anaerobic and live-in swampy soil or deep down in the soil (where water accumulates). It is the reverse of nitrification that occurs in the deep layers of soil where the bacteria convert NO_3^- into N_2 and other gaseous compounds like NO_2 . This occurs because in deep layers of soil, oxygen is not available and the soil bacteria use these nitrogen compounds instead of oxygen.

Thus, to conclude, microorganism play a central role in the nitrogen cycle as follows:-

- **Nitrogen-fixing bacteria**, which convert atmospheric nitrogen to nitrates.
- **Bacteria of decay**, which convert decaying nitrogen waste to ammonia.
- **Nitrifying bacteria**, which convert ammonia to nitrates/nitrites.
- **Denitrifying bacteria**, which convert nitrates to nitrogen gas.
- Fungi, like bacteria, help to convert dead plants and animals and their wastes into ammonia in the soil.

d. Microorganisms Give Plant Roots Excess to Nutrition in Soil

- Plant root creates a zone of nutrition depletion around their surface by using available nutrition.
- A group of fungi called mycorrhiza found associated with root hairs of plants helps in effective absorption of nutrition in soil.

(5) Bio-Control Agent

Biological control refers to the action of natural enemies like parasites, predators as well as pathogens in maintaining another organism's population density at a

lower level in nature than would occur normally in their absence. Biological method of controlling insect pest involves releasing predators/ parasites/pathogens which are the natural enemies of insect pests in sufficient number in crop fields. Conservation of natural enemies demands judicious and minimal use of insecticides on crops, so that parasites and predators are not unnecessarily killed. Selective insecticides which are not harmful to the natural enemies are used, such as organophosphates and methyl esters. Use of favorable application technique, e.g. soil application of systemic insecticides, seed treatment and use of baits, helps to conserve the natural enemies. Sometimes natural enemies are collected from the field, mass-bred in labs and then released in the field, much like biological insecticides, e.g. use of *Trichogramma* and *Bacillus thuringiensis*. Microbes that can control plant pests such as: *Bacillus thuringiensis* (BT), *Bauveria bassiana*, *Paecilomyces fumosoroseus*, and *Metharizium anisopliae*. These microbes are able to attack and kill various insect pests. Microbes that can control plant diseases such as: *Trichoderma sp.* capable of controlling plant diseases caused by *Gonoderma sp.*, JAP (white root fungus), and *Phytophthora sp.*

Biological control is a long-time self-perpetuating as well as economical method for controlling the insect pest. As these predators/pathogens are present naturally in the environment, there is absolutely no fear of pest developing resistance as seen with application of insecticides. Use of insecticides over a long period of time causes severe soil pollution. However, there is no such fear of environmental pollution with usage of natural predator/parasite population for controlling insect pest. This methodology helps in maintaining the ecological balance of the nature.

Also, with this methodology, there is no fear of pest resurgence, as typically happens with the application of insecticides. Biological control of insect pest is a time-consuming long-term process and often takes years before these natural enemies could be well established in crop fields. During this period, the insect pest can cause immense damage to the main crop. Sometimes, these natural predator/parasite population fails to establish successfully in the crop fields leading to failure of the entire programme. In case of sudden pest outbreak, bio-control fails to provide immediate relief. In a few cases, a natural enemy can also damage some other useful animals or plants. Bio-control is not 100 percent efficient and does not provide surety. Bio-control projects have equal chances of failure or success.

(6) Role of Microbes in Living Things

(a) The Process of Formation of Nodules on Legume Plant

Root nodules of leguminous plants like peas etc. are formed via a series of processes that preceded colonization *Rhizobium* bacteria on the hair root. *Rhizobium* bacteria colonization is thought to occur due to a plant protein known as “lectins” which might interact with specific *Rhizobium* bacteria that allow plants to recognize and accept the appropriate type of *Rhizobium*.

One *Rhizobium* strain can infect certain species of legumes; however it cannot infect others. For instance, the pea is the host plant to *Rhizobium*

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leguminosarum biovar viciae, on the contrary, clover acts as host to *Rhizobium leguminosarum biovar trifolii*. Specific kind of genes regulate the type of *Rhizobium* strain which can infects a particular kind of leguminous plant. Even if a strain is able to infect a legume, the nodules formed may not be able to fix nitrogen. Such rhizobia are called as ineffective. Effective strains induce nitrogen-fixing nodules. Effectiveness is ruled by diverse set of genes in the bacteria from the specificity genes. The different stages of nodulation are directed by the Nod genes. The initial interaction which happens between the host plant and free-living rhizobia involves the release of numerous chemicals by the root cells into the soil. A few enzymes positively affect the growth of the bacterial population in the area around the roots (the rhizosphere). Reactions occurring between certain compounds in the bacterial cell wall and the root surface are accountable for the Rhizobia knowing their accurate host plant as well as attaching to the root hairs. The nod genes in the bacteria are activated by Flavonoids. Flavonoids are secreted by the root cells. The activated nod genes induce the nodule formation. The whole nodulation process is regulated by highly complex chemical communications between the plant and the bacteria. Once bound to the root hair, the bacteria excrete nod factors. These stimulate the hair to curl. Rhizobia then invade the root via the hair tip where they can induce the formation of an infection thread. This thread is built by the root cells and not the bacteria and is formed only in response to infection. The infection thread grows via the root hair cells and enters other root cells present nearby. The bacteria then multiply extensively within the expanding network of tubes, continuing to produce nod factors which in turn stimulate the root cells to proliferate, leading to the formation of a root nodule. Within a week of infection, small nodules can be easily seen. Each root nodule is crowded with thousands of living *Rhizobium* bacteria, most of which are in the misshapen form known as bacteroids. Portions of plant cell membrane surround the bacteroids. These structures, known as symbiosomes, which may contain several bacteroids or just one, are where the nitrogen fixation takes place.

However, with the recent advent in technologies, symbiotic nitrogen bacterial preparations can already be purchased commercially. The farmers use such readymade symbiotic nitrogen bacterial preparations to inoculation seeds before planting beans. Though, between bacterial plant nuts have a certain degree of specificity, not all *Rhizobium* species produce nodulation and nitrogen fixation by any other legume crops. For the purpose of inoculation using commercial preparations of this bacterium, plant beans are divided into seven categories: alfalfa, “clover” (*Trifolium*), “pea” (*Pisum*), “vetch” (*Vicia*), “cowpea” (*Vigna*), “bean” (*Phaseolus*), lupines (*Lupinus*), and soybean. *Rhizobium* species or strains which are effective for other groups. Even within one species, certain strains are more effective than others against a particular host plant. Inoculation of seed before planting is an economical and efficient way to improve soil fertility and productivity. Most commercial preparations consist of selected strains of bacteria were added to the moist humus. Even then mixed with a certain amount of water and their distribution on the seed before planting.

(b) Symbiosis *Escherichia coli* in the Intestine Animals

Bacteria are not only present outside the animal’s body, but they also live inside the body too. This aggregate collection of microorganisms that is present in the

human body is termed as Microflora. *Escherichia coli* is referred to as one of the bacteria that cause intestinal disorders in humans. *E.coli* are short rod-shaped bacteria that can grow optimally at a temperature of 20-40 degrees Celsius. The presence of *E. coli* was first recognized by Theodor Escherich in 1885. *Escherichia coli* cause's diseases of digestive disorders, however, only a small fraction of this type of bacteria can actually cause potential damage to humans. The rest of the *E.coli* are beneficial in nature. *Escherichia coli* was found alive in the human colon wall and the function describing the remnants of food that is not absorbed in the digestive tract. This bacterium is also known to produce vitamin K and prevent the growth of other bacteria in the gut. Thus, bacteria are also present in the gut, and they aid the process of digestion by releasing certain enzymes. They live in a symbiotic relationship with a human. Other roles of microflora are vitamin K production, which is crucial in enabling blood clotting. They also prevent the invasion of the foreign bodies, by acting against other fatal microbes.

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(c) Allows Herbivore Animals to Get Nutrition From Poor Quality Food:

In ruminants' animals, the ingested food especially cellulose, lignin, etc., cannot be digested by the action of digestive juices in gut, however, the rumen (stomach) of these animals comprises of numerous bacteria, fungi as well as protozoa that can carry out fermentation where these complex carbohydrates are digested into simpler substances.

(7) Microbes Shape the Physical Structure of Soil

Soil's ability to store water and oxygen comes from its physical structure, which characteristically comprises of soil particles trapped together in stable formations known as aggregates. Microbes play an essential role in the development of these aggregates by releasing glue-like sugar substances that can stick soil mineral particles together and create new aggregates. A few advantages of having soil aggregate includes: - improved water infiltration rates, increased water-holding capacity, reduced soil compaction and reduced runoff. Proper soil structure also depends on having sufficient soil organic matter. Soil microbes also helps in the formation of soil organic matter. Many of the microbes in soils are what's known as decomposers. They have the ability to break down remains of dead & decaying organisms for the release of nutrients in the ecosystem. Further, when these microbes die, their carbon-rich bodies add to the soil organic matter.

(8) Recycling of Soil Nutrients

Microbes are active participants in soil nutrient cycling and thus play a key role in the storage, and eventual release, of plant-essential nutrients. For instance:- microbes like bacteria, fungi acting as decomposers have the ability to break down remains of dead & decaying organisms for the release of nutrients in the ecosystem. Thus, some of the nutrients locked in those tissues back into the soil, making them available for future plant consumption. These recycling cycles maintains a stable amount of plant-available nutrients in the soil.

This microbial recycling can also help growers use applied fertilizers more efficiently.

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(9) Microbes Strengthen Crop Health

Soil micro-biome can also contribute to the health of the crop. It can help to keep off pests and pathogens that can cause deadly plant diseases. Recent studies have suggested that some plants have become highly resistant to fungal disease due to their association with microbes present in soil. Further, it has been reported that soil microbes can also contribute to plant resiliency against other environmental stressors like drought. The researcher suggested that the bacteria were able to provide this benefit by producing special biofilms that increased crop drought resistance, and they are now looking into developing microbial amendments that can confer this drought resistance to more crops.

Thus, to conclude, the presence of microorganisms is very important and useful in maintaining the balance of nature. For that, we should learn and study deeper soil microorganisms so that the world of biotechnology in the field of microbiology can develop.

Check Your Progress

10. Which organisms are found in aquatic ecosystem?
11. How microorganisms play foundational roles in aquatic ecosystems?
12. Which organisms are considered as primary producers in the aquatic system?
13. Define Eutrophication.
14. What is Biological Oxygen Demand (BOD).

1.5 ANIMAL MICROBE SYMBIOSIS

Symbiosis refers to as an evolved interaction or close living relationship between organisms from different species, usually with benefits to one or both of the individuals involved. Symbioses interactions is of two types: -

- (a) **Obligate Symbiosis**- In 'Obligate' symbiosis, the two species are so interdependent on each other that either of the species is unable to survive without the other. Obligate symbioses are usually evolved over a long period of time.
- (b) **Facultative Symbiosis**- In 'facultative' symbiosis, the interacting species engage in a symbiotic partnership via choice, and can survive individually if needed. Facultative symbioses may evolve into obligate symbioses during the course of evolution.

Types of Symbiotic Relationship

The different types of symbiotic relationship are as follows:

(a) Mutualism

Mutualisms is a form of symbiosis wherein both the interacting species benefit from the interaction. This benefit can be in terms of fitness gain, strength gain, free food, free accommodation, etc. for either one or both the interacting species.

Thus, mutualisms can take the form of resource-resource relationships, service-resource relationships, or service-service relationships.

- Resource-Resource Mutualisms-** Resource-resource mutualism which is also referred to as ‘trophic mutualisms’ happen via the exchange of one resource for another between the two interacting species involved. Resource-resource kind of mutualisms most often occur between an autotrophic species, i.e., species which are capable of producing their own food from inorganic raw materials by the process of photosynthesis (basically a photosynthesizing organism) and a heterotrophic species, i.e., species which are dependent upon autotrophs for their food supply as they cannot synthesize their food directly using the solar energy (basically an organism which must absorb or ingest food to gain energy). Most of the plants have a trophic mutualism called a mycorrhizal association, which is a symbiosis between the roots of the plants and a fungus. The fungus colonizes the plants roots and is provided with carbohydrates, sucrose and glucose (free food) produced by the plants. In exchange, the plant benefits from the fungi’s higher water and mineral absorption capabilities.

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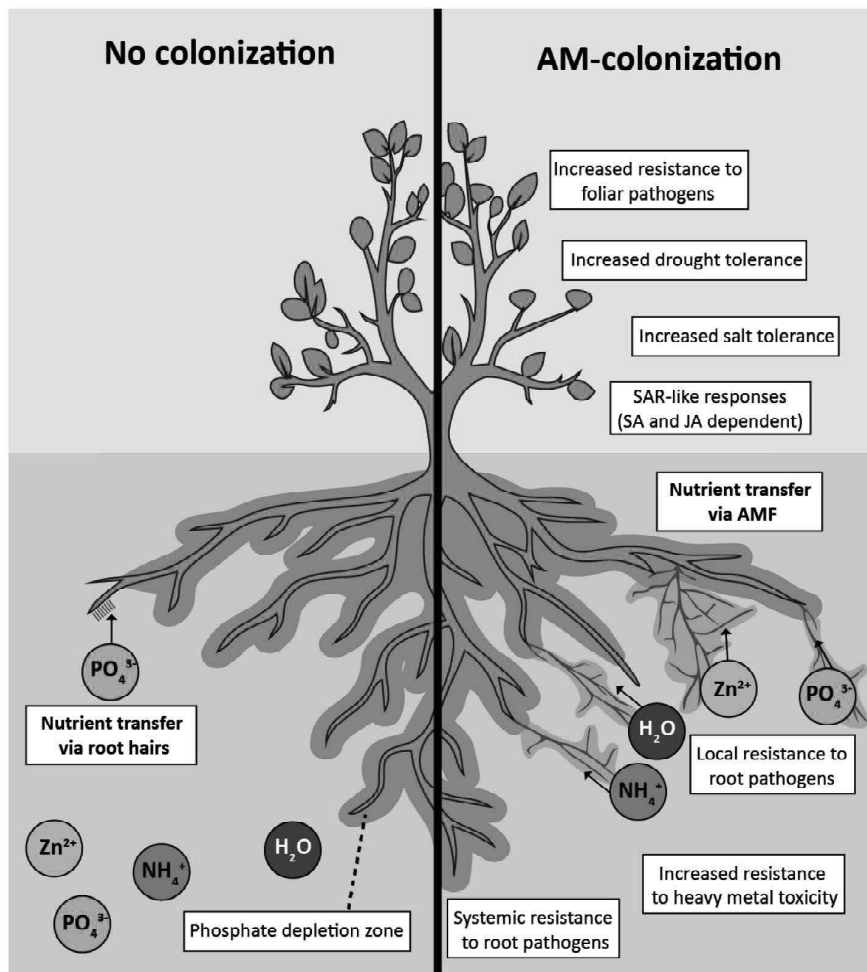


Fig.1.2 Symbiotic Association between Plant and Mycorrhizae

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Figure 1.2 is depicting the symbiotic association between plant and mycorrhizae. AM stands for Arbuscular Mycorrhizal Fungi [AMF]. They are soil fungi which form a mutualistic symbiosis with the roots of plants.

- **Service-Resource Mutualisms-** Service-Resource mutualism happens when the symbiotic species provides a service in exchange for a resource reward. A well-known instance depicting such symbiotic association is the exchange between plants and their pollinators. The pollinator (like insects, birds, moths, bats, etc.) visits the plants to gain a supply of energy-rich nectar. This provides the plant with the service benefit of being pollinated as their pollen grains gets attached to the hairs, antennae and other structures of insect while it is taking nectar from the plant. This ensures the widespread distribution of the pollen grains for plants whereas the insect gets the free energy rich food. This kind of service resource mutualism not only ensures the distribution of pollens but also the phenomenon of cross-pollination as described below:-

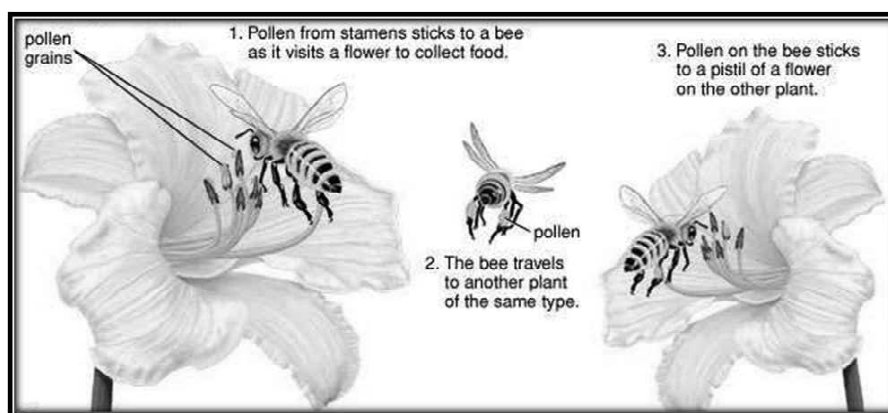


Fig. 1.3 Phenomenon Of Cross Pollination

Figure 1.3 is depicting the phenomenon of cross pollination.

- **Service-Service Interactions-** Service-service interaction is one of the rarest forms of symbiotic interactions. As the name suggests, both the interacting species receive a service, like shelter or protection from predators. For instance, the close association existing between the anemone fish (family: Pomacentridae) and sea anemones provides both the interacting species with protection from predators. The anemone fish, which have evolved an extra thick mucus layer on their skin to prevent them from being stung by the anemone's nematocysts, are provided with shelter from predators and a place to breed, while aggressively chasing away other fish which may try to bite the ends off the nutrient-rich tentacles. However, it is often said that there are only a very few truly service-service symbiotic mutualistic association as there is frequently a resource component to the symbiosis. In the case of the anemone-anemone fish mutualism, the nutrients from the anemone fish waste provide food for the symbiotic algae, which stays within the tentacles of the anemone and provide energy to the anemone via the process of photosynthesis. In this fashion, symbioses are shown to be highly complex and suggestive of the gentle balance that exist within the ecosystems.

(b) Commensalism

Commensalism refers to a symbiotic association where one species is benefitted from, and is often entirely dependent on the other for resources like food, shelter, or locomotion, with no noticeable adverse or beneficial effects on the host. The symbiotic association that exist between whales and barnacles is one of the well-known example of commensalism. The barnacles attach themselves to the tough skin of whales, and benefit from their widespread movement and exposure to currents, from which they feed, on the contrary, the whale is seemingly unaffected by the presence of barnacles.



Fig. 1.4 Commensalism between Whale and Barnacles

Figure 1.4 is depicting the commensalism between whale and barnacles

(c) Amensalism

Amensalism is contrary or opposite to commensalism. This happens when one species is inhibited or damaged by the presence of the other species. Amensalism may involve competition, where a larger, more powerful, or environmentally better adapted organism excludes another organism from its food source or shelter; for instance, one plant shades out another plant while growing at its normal speed as well as height. On the other hand, antibiosis, refers to the phenomenon where one organism secretes chemicals as by-products that kill or damage the other organism, but do not benefit the other, can be seen commonly in nature.



Fig. 1.5 Phenomenon of Amensalism

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Figure 1.5 IS depicting the phenomenon of amensalism. Here, algal blooms can lead to the death of several species of fish as well as other animals, however, the algae do not benefit from the deaths of these individuals.

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(d) Parasitism

Parasitism refers to the non-mutualistic form of symbiosis that occurs when one of the organisms benefits at the cost of the other. However, unlike predation, parasitism does not essentially/certainly lead to the death of the parasitized organism, and is usually, imperative to the life cycle of the parasite to keep its host alive. Occasionally, the parasitized host is killed due to parasitic invasion; in this case, the invader is referred to as a 'parasitoid'. Parasitism may involve direct infiltration of the host body to feed off tissue, influencing behaviours that benefit the parasite, or kleptoparasitism, in which the parasite steals food or other resources from a host. Parasitic symbiosis seems in numerous forms; few are rather non-threatening, for instance, ectoparasites, like fleas, which feed off the blood of larger animals and may cause uncomfortable itching. However, the ectoparasite may act as a carrier or vector, which transmits intercellular endoparasites such as bacteria and viruses to the host, often causing significant damage or death. Brood parasitism, which is a form of kleptoparasitism, can enforce a substantial cost on the fitness of the host. This has been commonly observed in birds (most notably in cuckoos), insects as well as some fish, where the parasite lays eggs within a host's nest, and later receives the resources like free food or free shelter intended for the host offspring. Brood parasitism can often lead to high numbers of offspring death, due to numerous reasons like starvation, rejection of the offspring or the abandonment of nests by the host parents, or by parasites removing host offspring from nests.

It has been hypothesized that a parasitic kind of symbiotic association might have developed/evolved from other, more giving/beneficial forms of symbioses; one species may begin to exploit a former mutualistic relationship by taking more of a resource or service than is returned, or by not contributing benefits to the other interacting species at all.



Fig. 1.6 Parasitic Association between Bed-Bugs and Human Skin

Figure depicting the parasitic association between bed-bugs and human skin.

1.5.1 Animal Microbe Symbiosis

As learnt from the different kind of interactions, symbiosis is the condition in which two different species stays together and exhibit spatial and temporal overlap in their interactions. The consequences of such interactions may be beneficial,

detrimental, or neutral; their nature often depends on ecological and environmental contexts and may result inter alia in mutualistic, parasitic, commensal, antagonistic, or neutral relationships. In this section, we shall discuss the different type of interaction existing between animal and microbes in nature.

(A) Beneficial Microbe Association with Sponges

A few sponges and beetles defend themselves against predators by making use of poisonous polyketide compounds produced by symbiotic bacteria.

(B) Association between Corals and Zooxanthellae

Corals are made up of animals known as corals polyps. Coral polyps bears highly specialized obligate mutualistic symbiosis with photosynthesizing algae known as zooxanthellae. They stay inside the coral tissue. The zooxanthellae are capable of capturing sunlight and converting the inorganic substance into organic substance (energy giving substance) with the liberation of oxygen. The organic energy producing substance are in the form of sugars and lipids that are transferred to the coral tissues and provide it with nutrients to survive and grow. In return, the zooxanthellae are provided with carbon dioxide, phosphorous and nitrogen as the by-product of the coral's metabolic process. This is one the finest example of mutualistic association. Even though, the corals cannot survive without any zooxanthellae, however, they can easily modify the number of zooxanthellae within their tissues, by altering the amount of nutrients that the algae receive. However, if the temperature of the water becomes too high for an extended period of time, the corals undergo stress and expel all of their zooxanthellae and are not provided with enough nutrients to survive. This results in coral bleaching. This kind of symbiotic relationship is called as endosymbiosis: a relationship in which one of the symbiotic species lives inside the tissue of the other.

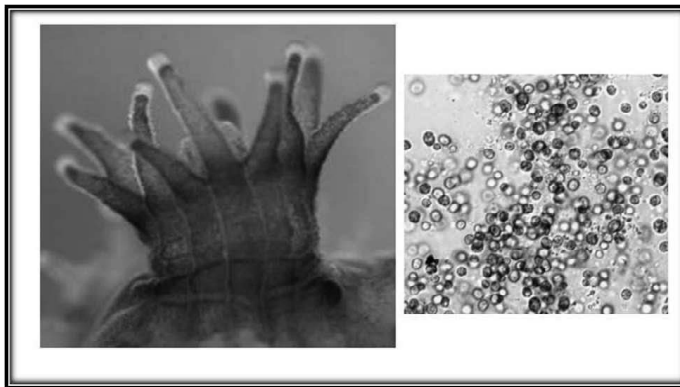


Fig 1.7 Association between Corals and Zooxanthellae.

Figure 1.7 is depicting the association between corals and zooxanthellae.

(C) Association between Termites and Trichonympha

Trichonympha is a genus of single-celled, anaerobic parabasalids of the order Hypermastigia that is found exclusively in the hindgut of lower termites and wood roaches. Trichonympha's bell shape and thousands of flagella make it an easily recognizable cell. The symbiosis between lower termites/wood roaches and

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Trichonympha is highly beneficial to both the interacting species: Trichonympha helps its host digest cellulose and in return receives a constant supply of free food and free shelter. Trichonympha possess numerous bacterial symbionts that are involved in sugar metabolism as well as nitrogen fixation.

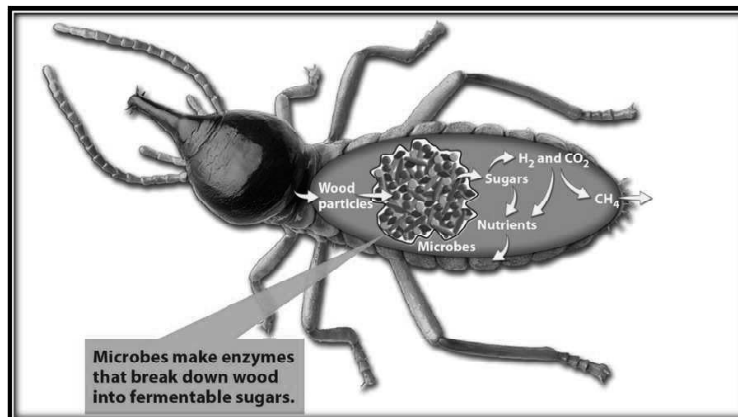


Fig. 1.8 Association between Trichonympha and Termite Gut

Figure depicting the association between Trichonympha and Termite gut.

(d) Beneficial Microbe Association with Arthropods

In aphids and many other insects, endo-symbiotic bacteria provide their hosts with nutritional components that are deficient in hosts' food sources. The majority of millions of insect species are associated with bacterial endosymbionts of the genus *Wolbachia*, which can affect host reproductive modes in a variety of ways.

An endosymbiont is any organism that lives either in specialized structures or inside any body part of another organism. Example includes nitrogen fixing bacteria which live in root nodules or several bacterial endosymbionts that provides essential nutrients to about 10% to 15% of the insects. In aphids and many other insects, endosymbiotic bacteria provide their hosts with nutritional components that are deficient in hosts' food sources. The majority of millions of insect species are associated with bacterial endosymbionts of the genus *Wolbachia*, which can affect host reproductive modes in a variety of ways. All types of interactions between insect and its endosymbiont can be categorized as symbiotic or pathogenic. On the basis of the extent of dependence, age of association and functional role played in the host; the endosymbionts can be categorized as either primary endosymbiont or secondary endosymbiont. All the primary bacterial endosymbiont share certain key features like vertical transmission, housing in specialized structure referred to as bacteriocytes and providing essential nutrients to the host.

Primary endosymbiont has established obligate relationship with the host and perform major functions like providing essential amino acids, vitamins, co-factors, storage and recycling of nitrogen as well as the factor required for fertility. In different insect species a variety of other S-symbionts also co-exists with P-endosymbionts. They are referred to as S-endosymbionts only when their intracellular location has been established. In aphid *A. pisum*, the rod-shaped, bacteriome sheath-associated S-endosymbiont was found to be related to the members of the Enterobacteriaceae. The possible functional role played by these

S-symbionts in host insect are ;rescuing host from heat damage, participation in host specialization and providing defence against enemies. They also play a major role in manipulating host reproduction. *Sodalis glossinidius* , the S-endosymbiont of *Glossina morsitans* has a genome size of 4171kb and a G+C content of 54.7%. It is the only s-endosymbiont whose genome has been sequenced. Here is an overview of the various insects and their primary endosymbiont.

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(1) Psyllid -*Carsonella*

Psyllid bacteriome has some structural variability. The most common bacteriome described consist of large multinucleate syncytiums within which are uninucleate bacteriocytes .The latter contain a polymorphic bacterium called *Carsonella ruddii*. The syncytial region of the bacteriome may also harbor a morphologically distinct S-endosymbiont. Use of specific oligonucleotide primer proved that *carsonella* is present inside bacteriocyte whereas secondary endosymbiont is present in the syncytial region.

Nucleotide determination of the three DNA fragments (37 kb in total) indicated that the G+C content of *carsonella* is just 20% and lacks intergenic spaces; most of the adjacent genes overlap using the sequence ATGA, in which the last triplet is the stop codon of the upstream gene and the first triplet is the start codon of the downstream gene.

(2) Whiteflies –*Portiera*

The orange coloured oval or roundish bacteriome (Buchner, 1965) of whiteflies contain *portiera aleyrodidarum* .In other members of the Sternorrhyncha the endosymbionts leave the bacteriocytes and enter the germ cells, in whiteflies the intact bacteriocytes migrate to the ovaries and enter the eggs.

Nucleotide analysis of the two DNA fragments 33kb in size revealed that G+C content of the DNA was 30.2 moles% (Baumann et al.,2004).

(3) Aphids-*Buchnera*

The bilobed bacteriome containing 60 to 80 bacteriocytes is present inside the body cavity of aphids. *Buchnera* is the common p-endosymbiot but in some cases it is replaced by yeast like endosymbiont.

The full sequence of genomes (616 to 642 Mb) was initially obtained for *Buchnera* of the aphid *A. pisum*, followed by *Buchnera* of the aphids *Schizaphis graminum* and *Baizongia pistaciae*. The genome of the *buchnera* is highly reduced, i.e., around 450 to 642 MB. The G + C content of these genomes are 25.3 to 26.3 moles %.

(4) Mealybugs-*Tremblaya*

Sap –sucking insects Mealybugs belongs to the family Pseudococcidae. They are referred to as Malybugs because they are usually covered with a mealy or cotton wax secretion. Within the body cavity of a female mealybug is a single large bacteriome composed of large bacteriocytes. The bacteriocytes contain vesicles within which is the P-endosymbiont, *Tremblaya princeps*. It has been reported that sometime *Tremblaya* contains an s-endosymbiont within its cells.

Nucleotide determination of two DNA fragments, 30 and 35 kb from *Tremblaya* revealed that G+C content (57%) of *Tremblaya* is higher than rest of the p-endosymbiont.

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(5) Sharpshooters-Baumannia

Sharpshooters are members of the suborder Auchenorrhyncha which feeds on xylem. As the composition of xylem is entirely different from the phloem so the function the p-endosymbiont in sharpshooters must be different from the p-endosymbiont of other members of Sternorrhyncha. The bilobed bacteriome of *Homalodisca coagulata* has a red-pigmented portion and a yellow-pigmented portion. P-endosymbiont is present within the red pigmented portion whereas a rod shaped bacteriome is present within the yellow pigmented portion. The genome size of *Baumannia* from the sharpshooter *H. coagulata* is 680 kb. The G + C content of a 3-kb endosymbiont DNA fragment encoding for *rpoBC* is 37 moles %.

Functions of Endosymbionts

(1) Contribution of Microbiota to Nutrition

The dietary contribution of gut microbiota in insect host includes production of essential amino acids, vitamins and release of digestive enzymes which enables the insect to survive and flourish on nutritionally imbalanced or suboptimal diet as described above. Hemipteran insects like aphids, white fly, plant hoppers, mealy bug, etc. feed exclusively on plant sap. which is rich in carbohydrate but lack essential amino acids and nitrogen. Primary obligate endosymbionts like *Portiera* in white fly or *Buchenra* in aphid provides all these essential amino acids required by the insect. Members of the family Enterobacteriaceae like *Pantoea septica* and *Erwinia chrysanthemi* were reported to be potential diazotrophs and thus supplement the host diet with essential amino acids. In case of termites and fruit fly, gut bacteria has been reported to play an essential role in nitrogen fixation.

(2) Contribution of Microbiota to Colonization Resistance

The indigenous gut microbiota of an insect provides an effective resistance against colonization by pathogenic bacteria or fungal communities—a mechanism referred to as colonization resistance. It was found that axenic locusts were susceptible to fungal infections as compared to locust with healthy microbiota. Analysis of the gut content of the conventionally raised locust confirmed the presence of phenolic compounds that were antifungal in nature. Monoassociation of locust with *Pantoea agglomerans* led to partial production of antifungal phenolic compounds. Experiments conducted in axenic silkworm larvae exhibited that their gut was susceptible to colonization by pathogen like streptococci and *Serratia piscatorum* compared to the conventionally raised larvae. *Hamiltonella defensa*, a secondary endosymbiont of aphids, has been shown to protect its host from parasitoid *Aphidius ervi*. Further, *Wolbachia* has been proven to augment its host fitness by providing protection against parasitisation.

(3) Contribution of Gut Bacteria in Reproduction

In class Insecta, *Wolbachia* is almost ubiquitously present in several orders including Hemiptera, Lepidoptera, Diptera, Coleoptera and Hymenoptera (Hoerauf and

Rao, 2007). Wolbachia can be transmitted both horizontally (though less commonly) and vertically from parent to offspring via female's eggs. Wolbachia has been reported to induce reproductive manipulation in insects by mechanism like cytoplasmic incompatibility for its successful transmission from one generation to another. In *Drosophila* spp., it has been shown that uninfected (lacking Wolbachia) females failed to produce offspring when mated with Wolbachia infected males, whereas infected females (having Wolbachia) produced viable offspring when mated with either Wolbachia uninfected or infected males. Thus, infected females were more reproductively fit as compared to uninfected females. In some insect species, Wolbachia infected females can reproduce parthenogenetically. The resulting offspring were all females and also carried the same Wolbachia strain. Wolbachia has also been reported to suppress the production of male hormones causing all the fertilized eggs to develop into females irrespective of their genotype. In some insect species, it also caused the males to abort early, thus allowing only the female to develop.

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(4) Gut microbes produce semiochemicals that play an important role in insect behaviour

In *S. gregaria*, it has been observed that the majority of the pheromone that affects insect behavior was phenolic compounds like guaiacol and phenol released from fecal pellets. Monoassociation of gnotobiotic locust with the bacterium *P. agglomerans* led to the production of these two phenolic compounds. Further, an in-vitro study has shown that *P. agglomerans* was capable of producing both these compounds from fecal pellets of locust.

(5) The myrmeleontidae larvae can easily survive on the body fluids of its prey for one to three years. *Enterobacter aerogenes*, residing in host salivary glands, produces a homologue of GroEL chaperone that acts as toxin and helps to paralyze the prey.

(6) N-acylamino acids are the oral secretion of an insect that triggers the production of volatile compounds in plants. These volatile compounds draw predators towards herbivores. In *S. littoralis*, gut bacteria were involved in the production of N-acylamino acids. This suggests towards possible induction of another trophic level among insects, its plant hosts and predators.

(7) Excessive use of pesticides has not only deteriorated the environment but its continuous use has made the insect highly resistant to majority of the insecticides and pesticides. Modern integrated pest management strategies employ host microbial communities to control pests. In a study conducted by Ateyyat et al., the potential of eleven bacterial isolate was studied as effective biological control agent towards whitefly. Results suggested that three bacterial isolate, i.e., *Erwinia persicinus*, *Bacillus pumilus*, and *Exiguobacterium acetylicum* were proved effective and have shown significantly high mortality level against 2nd instar whitefly nymphs.

8) In Lepidopterans, *Enterobacter* spp. has been shown to aid in the toxicity of *Bacillus thuringensis* (Broderick et al., 2006). A few species of *Enterobacter* was reported to be mildly pathogenic and release prodigiosin- an antiparasitic compound.

9) In *Acyrtosiphon pisum*, infection by facultative symbiont *Serratia symbiotica* or *Hamiltonella defensa*, provide tolerance to high temperature.

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Gut-Bacteria in Humans

Microbiota refers to a group of microorganisms that exist in a commensal interaction with the host surface as they colonize a particular tissue surface. The system formed is known as normal microbiota, and it majorly comprises of bacteria, viruses, fungi, protozoans etc. All these microbial species exist in a commensal relationship with the host species and depend on the host for nutrition as well as shelter. Although, the diversity of microbiota grows in close association with the host, they do not cause any ill effects or harm to the host body. Though, in few situations, in individuals with reduced immunity and immune-compromised conditions, these species might cause different diseases especially gut disorders like irritable bowel syndrome. Gut bacteria is especially significant for digestion in humans as well as other species. In human beings, gut bacteria contribute in breaking down the additional carbohydrates, out-competing the disease-causing bacteria and producing special hormones to direct fat storage. Humans lacking healthy mutualistic gut flora can suffer a variety of gut disorder like irritable bowel syndrome. Further, in humans, a diverse group of microorganisms resides in different parts of the body, but the most diverse and the largest population of such organisms reside in the gut. Different microbes are known to exist on the surface of human tissue and depend on the host for nutrients as well as living space. However, it should be noted, that even though the healthy microbiota resides in the gut system and takes up nutrients from the digestive system, yet, it doesn't cause any harm to the host unless the species is pathogenic. In few situations, the microbial species might be opportunistic pathogens and cause gut disorder in immunocompromised individuals. Dysbiosis refers to an undesirable shift in the microbiota composition, leading to an imbalance between protective and potentially harmful microbes that can damage the gut lining and lead to chronic diseases. *E. coli* is a facultative anaerobe that utilizes oxygen and lower the oxygen concentration in gut which creates suitable environment for obligate anaerobes such as *Bacteroides*. *E. coli* is a host which remains unaffected by *Bacteroides*. Lactic acid produced by many normal floras in vaginal tract is inhibitory to many pathogenic organisms such as *Candida albicans*. Fatty acid produced by skin flora inhibits many pathogenic bacteria in skin

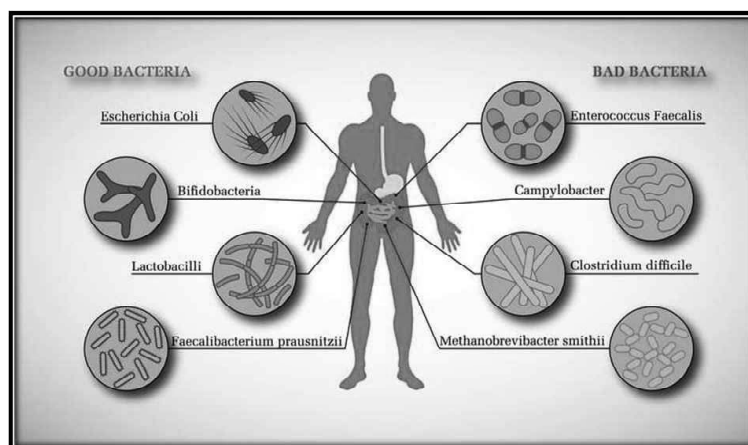


Fig. 1.9 Good Bacteria and Bad Bacteria in Human Being

Figure 1.9 is depicting the good bacteria and bad bacteria in human being.

(f) Gut Bacteria in Ruminants

Ruminant animals, such as cows or deer, depend on special mutualistic bacteria which help them to break down the tough cellulose in the plants they eat. In return, the gut bacteria get a stable free supply of food. Ruminant animals (like deer and cows) digest food in a four-chambered stomach with the help of special microbiota like bacteria, protozoa as well as fungi. Ruminant animals utilize a special four-chambered stomach with a unique microbial flora to digest tough cellulose found in the plants in their diets. Most vertebrates cannot make cellulase, the enzyme that breaks down cellulose, but microbes in the rumen produce it for them. Ruminants are of special interest to microbiologists as they have specialize/unique species of bacteria, yeasts, protozoa, and fungi in their rumens. The plant matter consumed by ruminants is very high in cellulose content, however, vertebrates cannot produce cellulase which is the enzyme required to break down the cellulose. Thus, ruminants depend on the symbiotic microbes in their guts to break down cellulose for digestion. In the absence of oxygen in the rumen, bacteria in the rumen are usually anaerobes or facultative anaerobes.

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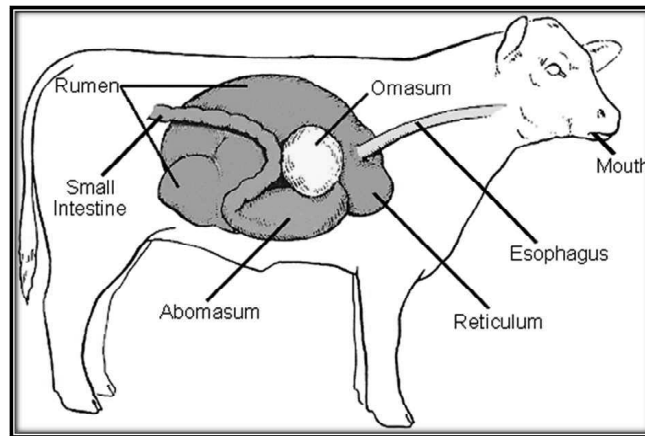


Fig. 1.10 Digestive Tract of Ruminant Animal

Figure depicting the digestive tract of ruminant animal.

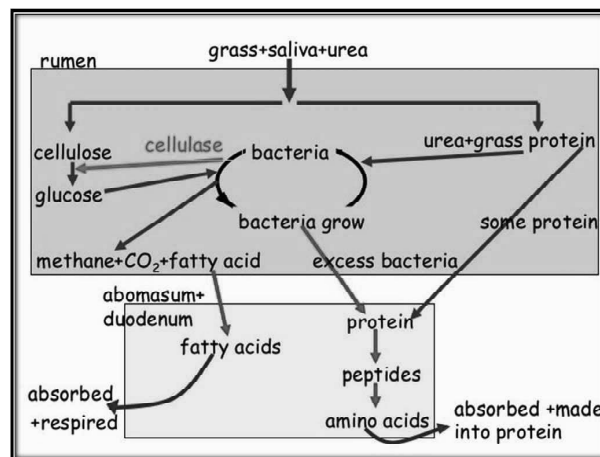


Fig. 1.11 Digestion in the Rumen of Digestive Tract

Figure 1.11 depicting the digestion in the rumen of digestive tract.

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(g) Animal Microbe Association in Hydrothermal Vents

Hydrothermal vents emit nutrient rich, geo-thermally heated water. Mats of chemosynthetic bacteria grow and flourish around the vents and synthesize carbohydrates from the carbon dioxide ejected by the vent. Many animal species of crabs, worms, snails, and tube worms depend on these bacterial mats for their food supply. These species are often specially adapted to life in the lightless, high pressure, and hot environment of the vent. Vents are the prime target of the mining industry causing exploitation of resources. This has become a major cause of concern among marine biologists. Mining activities could potentially damage these very unique and diverse ecosystems.

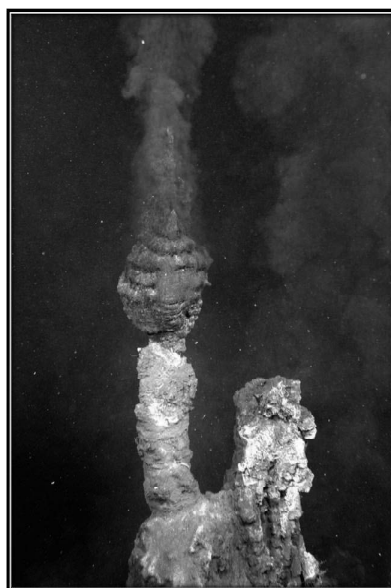


Fig. 1.12 Hydrothermal Vent

Figure 1.12 is depicting the Hydrothermal Vents. Hydrothermal vents are cracks in the earth's crust where geothermally heated water leaks out. In most of the shallow water as well as terrestrial ecosystems, energy comes from direct sunlight, however, in the deep ocean there is total darkness as sunlight is not able to penetrate deep. Thus, in the absence of direct sunlight, hydrothermal vents often expel nutrient rich water, containing methane and other sulfur rich compounds. A hydrothermal vent refers to a fissure in the earth's surface from which geothermally heated water issues. They are frequently present deep below the surface of the ocean. Hydrothermal vents are of great interest to microbiologists as they have unique microbial communities found nowhere else on earth. Vent bacteria are capable of synthesizing all the organic compounds they need to live from these nutrients, a process referred to as chemosynthesis. Chemosynthesis refers to the production of carbohydrates as well as other organic compounds from simple inorganic compounds like carbon dioxide, utilizing the oxidation of chemical nutrients as a source of energy rather than sunlight; it is limited to few species of bacteria and fungi. These bacteria form the basis of the entire hydrothermal vent ecosystem. The chemosynthetic bacteria grow into a thick mat, covering the hydrothermal vent, and this represents the first trophic level of the ecosystem. Snails, shrimp

crabs, tube worms, and fish feed on the bacterial mat and attract larger organisms like squid and octopuses. Majority of these species are particularly adapted to live in the dark and hence lack eyes. Hydrothermal vents are biodiversity hot spots as they have numerous species that are uniquely adapted to survive in this harsh environment. For instance, the Pompeii tube worm *Alvinella pompejana* can resist temperatures up to 176°F. These ecosystems are almost completely independent of sunlight (although the dissolved oxygen used by some animals does ultimately come from plants at the surface).

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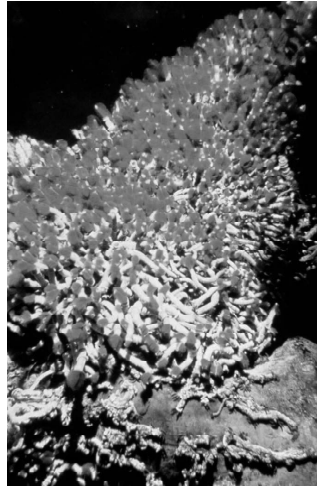


Fig. 1.13 Tubeworms Residing Near a Hydrothermal Vent

Figure is depicting the tubeworms residing near a hydrothermal vent. Few species of tube worms are particularly adapted to resist/tolerate the high temperatures found at hydrothermal vents.

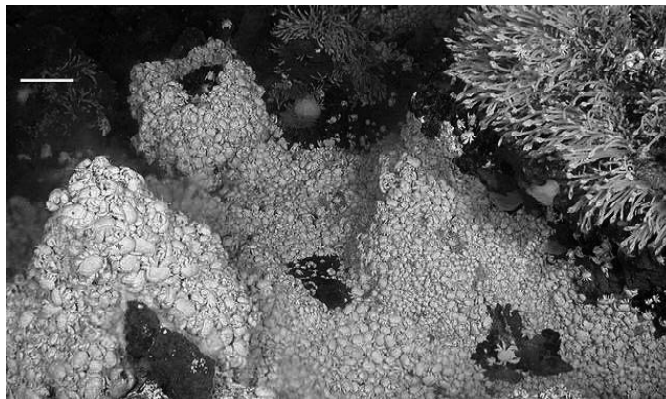


Fig. 1.14 Crabs Residing Near a Hydrothermal Vent

Figure 1.14 depicting the crabs residing near a hydrothermal vent. The ecosystems around hydrothermal vents depend on mats of chemosynthetic bacteria, and numerous species feed on the bacteria. Hydrothermal vents are some of the most unique ecosystems in the world

Even though, hydrothermal vents are one of the most remote ecosystems in the world, they are under threat from mining companies. Due to depletion of mineral resources from earth surface, mining companies have turned to deep sea geothermal

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vents to extract metals as well as other sulfur compounds. Even though, the technology for deep sea mining is new, conservation biologists are majorly concerned that mining hydrothermal vents will destroy these fragile and unique ecosystems.

(h) Bioluminescence

A special category of symbiotic interactions involves bioluminescence. Here, light producing bacteria are hosted by another organism. A well-known example of bioluminescence is the Hawaiian bobtail squid (*Euprymna scolopes*) and its mutualistic bacteria, *Aliivibrio fischeri*. *Aliivibrio fischeri* inhabits a special light organ in the squid's mantle. The bacteria are fed upon a diet rich in sugar and amino acid by the squid. In return, the bacteria produces, light to hide the squid's silhouette when viewed from below, allowing the squid to match ambient light conditions.



Fig. 1.15 Bobtail Squid

Figure 1.15 depicting the Bobtail Squid: Bobtail squid depend upon their mutualist bacteria *Allivibrio fischerii* to generate light. The bacteria inhabits in a specialized light organ having ciliated cells in the squid's mantle and receives sugars and amino acids in exchange for light.

However, the hatchlings of Bobtail squid do not possess *Aliivibrio fischeri* naturally in their bodies. They are born with a specialized light organ structure, having ciliated cells at the opening intended to trap the passing *A. fischeri*. Thus, the bacteria are obtained from sea water. To obtain bacteria from sea water, the squid secretes a special kind of mucus whenever its cells detect peptidoglycan (a special complex protein-carbohydrate complex which is found in the cell walls of bacteria). The mucus secreted by squid collects near the opening of the light organ which then traps the passing bacteria. The squid weeds out unwanted bacteria in numerous ways. For instance, *A. fischeri* is able to survive in the mucus better when compared to other bacterial species. *A. fischeri* is also a very mobile bacteria, and is able to swim against the water current created by the cilia lining the mouth of the light organ. Further, squid secretes an enzyme at the entrance of light organ. This enzyme is capable of splitting hydrogen peroxide, thus creating a toxic environment for most of the bacterial species. However, *Aliivibrio fischeri* can

capture hydrogen peroxide before the squid can use it as a toxin, and thus can survive in the hostile chemical environment. In this way, squid makes sure that only light producing bacteria get access to the light organ. Once, *A. fischeri* has passed through all these barriers at the opening of the light organ, it can conveniently colonize the chambers of the light organ and begin enjoying the benefits of symbiosis. Despite all the effort that goes into obtaining *Aliivibrio fischeri*, the squid ejects 95% of its bacteria every day. However, it not clearly known that why the squid cleans out its light organ. However, as the bacteria require a great amount of sugar as well as amino acids, hence, it might be most useful to the squid to host bacteria only when they are needed. It may also provide a supply of bacteria for squid hatchlings.

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Plant Microbe Symbiosis: Mycorrhizae

The roots of most species of seed plants (at least 80%) have a symbiotic relationship with soil fungi in which both organisms benefit. The associations are known as **mycorrhizae** (sing : mycorrhiza).

Part of the fungal mycelium is in the host tissue, and part is in contact with the surrounding soil and usually spreads into it. Mycorrhizal associations increase the area of contact of the roots with the soil. The extension of hyphae into the soil also makes the soil more stable. It is likely that the great majority of land plants enter into this kind of relation with soil fungi and sometimes the host cannot thrive without the benefits derived from the fungus. They are of great significance because they are probably the major route of entry of water and nutrients into roots especially when the plant is growing in nutrient – limiting soils.

A very diverse array of mutualistic associations exist between the fungi and the roots of plants. The fungus receives organic nutrients, mainly carbohydrates and vitamins from the plant and in return absorbs mineral salts (particularly phosphate, ammonium, potassium and nitrate) and water which can pass to the plant root. Generally only young roots are infected. Root hair production either ceases or is greatly reduced in infection. Host growth is almost always enhanced compared to uninfected plants of the same species.

Mycorrhizae also may be beneficial to their host plants by secreting hormones or antibiotic agents that reduce the potential of plant disease.

Mycorrhizae occur in some bryophytes (especially liverworts), many pteridophytes, all groups of gymnosperms, and nearly all families of angiosperms. In the families, e.g., Juncaceae, Cyperaceae, Caryophyllaceae, Chenopodiaceae and Cruciferae, only a few species are regularly infected. Fungi that are involved in the mycorrhizal associations include representatives of zygomycota, accomycota, basidiomycota and deutermycetes.

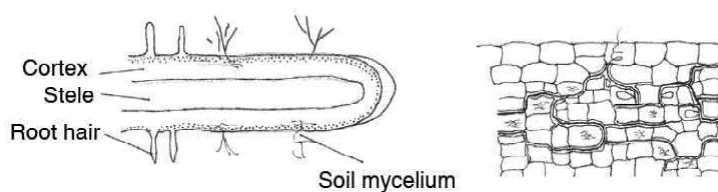
On morphological and anatomical features such mycorrhizas can be divided into four principal types:

1. Vesicular-Arbuscular Mycorrhizas (VAM)

They are by far the commonest of all mycorrhizas and are found in Bryophytes, Pteridophytes, Gymnosperms (excluding the Pinaceae which have sheathing

mycorrhizas) and virtually in all families of angiosperms (Ericaceae and Orchidaceae have other types of mycorrhiza).

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A L.S. of root let showing fungal penetration

B T.S. root cortex showing 'Arbuscules' and 'Vesicles' formed by the infecting fungus

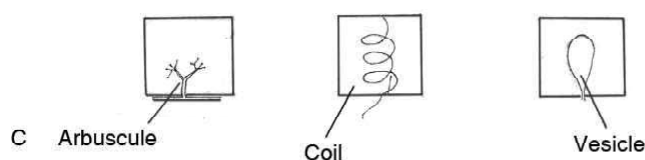


Fig. 1.16 Vesicular-arbuscular mycorrhiza. A—L.S. of rootlet showing fungal penetration B—T.S. root cortex showing external hyphae, hyphal coils, intercellular hyphae, vesicles and arbuscules in various stages. C—cells of a rootlet showing host/symbiont contacts.

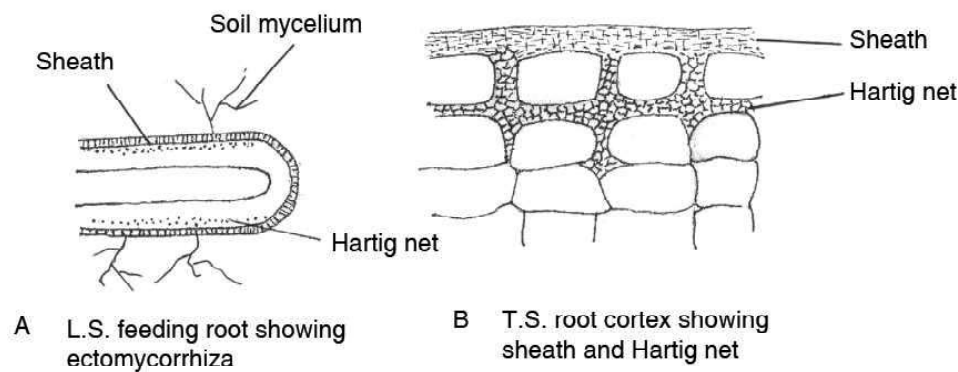
The name vesicular arbuscular derives from structures formed by the infecting fungus in host tissue. Fungus hyphae invade the cell but do not break the host plasmamembrane. Inside the cell they branch repeatedly forming dichotomously branched projections called an **arbuscule**. **Vesicles** are terminal or sometimes intercalary swellings which may either be between host cells or within them. Vesicles have been ascribed a storage and reproductive function. Within the plant, the mycelium develops only in primary cortex and epithelium of the root and does not penetrate the endodermis into the conducting tissues within the stele. Hyphae within the root are connected to an external soil mycelium. The infection does not alter the morphology of the root or its external appearance and root hairs may be formed.

The group of fungi forming VAMs belong to a single family, the Endogonaceae of Zygomycotina.

2. Ectomycorrhizas

Ectomycorrhizas are believed to be more common in temperate zones of the world, where there are seasonal climatic changes, than in the tropics; these mycorrhizas are mainly formed on roots of woody plants.

In such mycorrhizas, the ultimate absorbing rootlets of the root system are completely surrounded by a distinct **mantle** or **sheath** of fungal tissue (Figure 1.17) which also encloses the root apex. Fungal hyphae extend inwards from the sheath into the outer 2-3 cell layers of the cortex forming an intercellular network of hyphae known as the "**Hartig net**".



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Fig. 1.17 Ectomycorrhiza. A—L.S. infected rootlet showing the sheath and Hartig net, B—cross-section of a rootlet showing fungal sheath and intercellular hyphae.

Fungi which form ectomycorrhizas include several genera of Gasteromycetes of Basidiomycotina, 18 Ascomycetes and Zygomycete genus *Endogone*.

3. Ericaceous Mycorrhizas

The order Ericales are mostly woody shrubs or trees and form ecologically important plant communities particularly on moors, swamps and on peat (nutrient-poor soils).

Three different types of mycorrhizas are formed by the Ericales.

(a) **Ericoid mycorrhizas:** These occur on plants which have very fine, hair-like roots and cortex is composed of only a few cells. In the mature phase of the infection, every cell of the cortex is filled with extensive **coils** of hyphae. There are numerous hyphal connections between the root and soil surface (Figure 1.18).

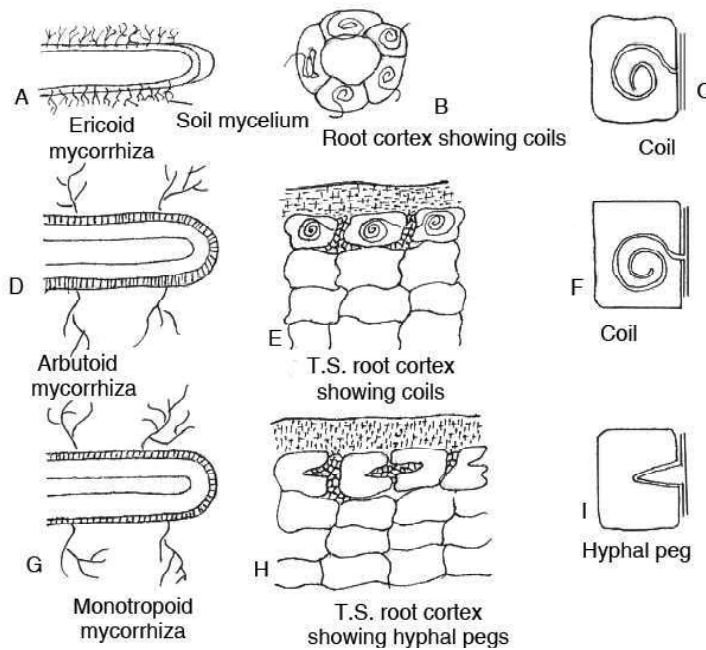


Fig. 1.18 Ericalean mycorrhizas. A-C—Ericoid mycorrhiza. D-E—Arbutoid mycorrhiza, and G-I—Montoropoid mycorrhiza.

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(b) Arbutoid mycorrhizas: These occur on the members of the Arbutioideae and Pyrolaceae.

The roots are infected in a manner resembling ectomycorrhizas in that they are enclosed in a fungal sheath. A Hartig net is also formed, but this differs from that of ectomycorrhizas by its restriction to the outer epidermal layer and by abundant penetration into cells by coils of hyphae.

(c) Monotropoid mycorrhizas: *Monotropa* plant is usually herbaceous and lacks chlorophyll. They have mycorrhizas which facilitate transfer of carbon compounds from the roots of photosynthetic host plants.

Monotropoid mycorrhizas resemble arbutoid in having a sheath and a Hartig net limited to the epidermal layer, but differ in that the penetration into host cells are in the form of simple **pegs** (Figure 1.18).

4. Orchid mycorrhizas

All orchids are infected with fungi at an early stage of their life, when the entire young plant is still underground and non-photosynthetic. As in monotropoid mycorrhizas, the plant receives carbohydrate from the fungus.

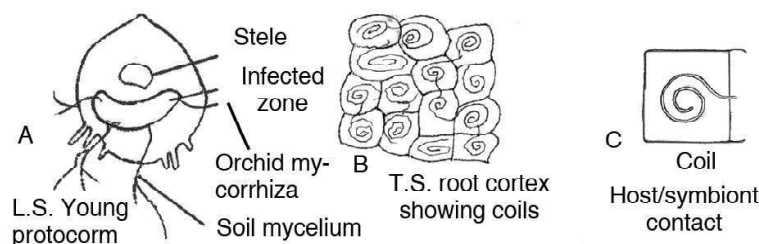


Fig. 1.19 A-C—Cross section of an orchid mycorrhiza showing fungal penetration and coils of hyphae

Infection occurs after the seed coat has been ruptured. The fungal penetrations develop into coils (Figure 1.19). The mycelium spreads within the plant, invading the cells as it grows.

Check Your Progress

15. What do you understand by symbiosis?
16. Define service-service interaction.
17. What is commensalism?
18. What is the name given to any organism that lives either in specialized structures or inside any body part of another organism?
19. Define mycorrhizae.
20. What is the benefit of mycorrhizae to the host plant?

1.6 MICRO-ORGANISMS IN FORMATION OF DIFFERENT SOILS

Soil may be defined as “the part of earth crust in which humus is present”. According to R.F. Daubenmire, “soil is anchored.” He defines soil as weathered superficial layer of earth crust with which are mingled living organisms and products of their decay. According to Hilgard, 1917 (American school), “it is, more or less, loose, friable material in which, by means of their roots, plants may or do find a foothold, nourishment as well as other conditions of growth.”

According to Raman, 1928 (German school), “soil is the upper weathering layer (i.e., layer subjected to physical and chemical changes) of the solid earth crust.”

Joffe and Marbut, two well-known American soil scientists, have defined soil in the following way: “soil is a natural body developed by natural forces acting on natural materials. It is usually differentiated into horizons of minerals and organic constituents of variable depths which differ from the parent materials in morphology, physical constitutions, chemical properties, composition and biological characteristics.” According to Russian school, “soil is natural body differentiated into horizons of usually unconsolidated minerals and organic constituents of variable depths.” According to Wadia (1945), “soil is the topmost layer of earth crust capping the rock.” It is natural body of variable thickness, composed of disintegrated rock materials together with variable proportions of organic matters, generally differentiated into zones or layers and mostly unconsolidated. In brief, soil can be defined as that region on the earth surface where geology and biology meet each other.

The soil is made up of the following components:

- (1) Mineral particles,
- (2) Dead organic matter or humus,
- (3) Soil atmosphere;
- (4) Soil water, and
- (5) Biological system or soil micro-organisms.

Mineral Components

The mineral constituents of the soil are derived from the parental rocks or regolith. They may be found in the form of particles of different sizes; from clay (.0002 mm or less in diameter) to large pebbles and gravels. The minerals represent about 90% of the total weight of the soil. Important elements which are found in compound state are Oxygen, Si, Fe, Al, N, P, K, Ca, Mg, C, H, etc. In soil, nitrogen comes from atmosphere in the form of nitrogen salts.

Organic Matter or Humus

Besides inorganic minerals, some organic residues derived either from dead remains of plants and animals or through metabolic activities of living organisms are present in the soil. When the plants and animals die, their dead remains are acted upon by a number of microorganisms and are finally degraded or decomposed into simple

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organic compounds. A product of this microbial decomposition is humus which is a dark coloured, jelly-like amorphous substance composed of residual organic matters not readily decomposed by soil microorganisms. The process of humus formation is called humification. The chief elements found in humus are carbon, hydrogen, oxygen, sulphur and nitrogen. The important compounds found in it (humus) are carbohydrates, phosphoric acid, some organic acids, fats, resins, urea, etc. Tree litter (very little decomposed dead matter) also contains some inorganic substances as lime, potash, Mn, Mg, silica, Cu, Al, Ca, Na, K, etc. Humus is a dynamic product and is constantly changing because of its oxidation, reduction and hydrolysis. Hence, it has no definite chemical composition. It has much carbon content and less nitrogen.

Humus is not soluble in water. It is present in soil in the form of organic colloids. The amounts of humus in different soils vary greatly. Humus percentage in the soil is affected by climatic and biological factors. It is less in arid soils and very high in humid soils. In the top layer of the soil, humus quantity is greater than in the deep layers. In dark humid areas which are thickly covered with vegetation, the humus may be found in the following three stages of degradation:

1. The top floor is covered with dead organic parts showing low degree of decomposition. These poorly decayed dead parts of plants form litter.
2. Below the litter may be found a layer of partially decomposed organic matter which is known as duff layer.
3. When the duff is decomposed completely into organic substances, the decomposition products, generally called leaf moulds, are accumulated below duff layer.

Biological system of the soil or soil microorganisms

Organisms present in the soils are called soil organisms. Important group of soil organisms are given below:

Soil Flora

1. Bacteria
2. Soil fungi
3. Soil actinomycetes
4. Algae
5. Root, rhizoid and rhizome of higher plants

Soil Fauna

1. Protozoa
2. Nematodes
3. Insects and mites
4. Rodents and earthworms
5. Burrowing vertebrates

Algae are found in the top layer of soil under the conditions of constant shade and moisture. It is estimated that in soil microflora bacteria form about 90 per cent of the total microbe population. Fungi and algae together represent only

one per cent and actinomycetes cover 9 per cent. Density of microbial population is actually governed and influenced by climatic conditions, physical and chemical nature of soil and vegetation cover. The greatest amount of microbes (10,00,000 per cubic cm) is found in the top layer of soil at a depth of 5 to 15 cm. In deeper layer (1.5 to 5 m) individual microbes are found. However, they have been discovered at a depth of 17.5 m in coal, oil and artesian water. It has been calculated that in the ploughed layer of cultivated soil over an area of one hectare there may be from 5 to 6 tons of microbial mass and one gram of ploughed soil contains 1-10 thousand million bacteria.

Role of Soil Organisms in Formation of Soil

Soil organisms take part in a number of processes in the soils. Some of their important roles are as follows:

- (1) Decompose the dead organic matter and increase plant nutrients in available forms,
- (2) Production of toxins,
- (3) Production of growth stimulating substances,
- (4) Nitrogen fixation in the soil,
- (5) Mixing of soil,
- (6) Improvement in soil aeration,
- (7) Improvement in the aggregation of soil particles or soil binding, and
- (8) Cause injury to the plants.

Remains of plants and animals and cause decomposition. In the process of decomposition, complex organic matters are converted into simple organic compounds. Compounds like sugars, starch and proteins are decomposed first in the decomposition process and then cellulose, fatty substances and lastly lignin and woody substances are degraded. Proteins when acted upon by microbes are converted into amino acids, ammonium salts, nitrates and nitrites. Humus, an intermediate product of decomposition process, is formed by micro-organism in optimum physical conditions. In the decomposition process, a number of complex mineral compounds are also converted into simpler and soluble compounds.

1. Organic acids and carbon dioxide that are released by decomposition make insoluble phosphates and other unavailable compounds more easily available to plants. Decomposition of dead organic matter primarily helps in the feeding and growth process of these micro-organisms and secondly, increases the nutrient contents of the soil. Bacteria and soil fungi are main agents which bring about the process of decomposition in the soil.
2. Production of toxins. In the absence of oxygen some soil microbes secrete chemicals, such as, aldehydes, organic acids, etc. which may show toxic effects on many plants. Examples of toxin secreting organisms may be found in fungi, bacteria and algae. *Fusarium lini*, which causes wilt of flax (*Alasi*) secretes HCN, a deadly poisonous substance and *Fusarium udum*, a fungus causing wilt of pigeon pea (*Arhar*) secretes fusaric acid in the roots of the host plants. These toxic chemicals secreted by fungi may be responsible for causing wilt in the flax and arhar (*Cajanus cajari*).

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3. Production of growth stimulating substances. Many soil organisms including soil fungi and bacteria produce growth stimulating substances such as 3-Indol acetic acid, Gibberellins and Gibberellic acid in the soil. *Fusarium* species too have been found to secrete Gibberellin and Gibberellic acid (C₁₉H₂₂O₆).
4. Nitrogen fixation. Many bacteria inhabiting the root nodules of leguminous plants (*Rhizobium*), nitrogen-fixing bacteria living free in the soil (*Azotobacter*, *Clostridium pasteurianum*), actinomycetes, fungi, purple bacteria and a number of blue-green algae are known to fix free atmospheric nitrogen gas into nitrogenous compounds, such as nitrates and nitrites, etc., and thereby increase the fertility of the soil. It has been established that in each hectare of ordinary soil every year 25—50 kg of nitrogen are fixed and in cultivated soil and in soil containing legume plants 35 to 60 kg and 100 to 400 kg of nitrogen are fixed respectively. *Anabaena*, *Nostoc*, *Microcystis* are important nitrogen fixing blue-green algae. De and Fritsch (1938) have found that certain blue-green algae are able to fix 20 lbs of atmospheric nitrogen per acre in a rice field. They increase the yield of rice from 15 per cent to 25 per cent. Singh, R.N. and Relwani and others have also shown experimentally that some blue-green algae fix nitrogen in the paddy soils.
5. Soil mixing. Many organisms by their mechanical activities help in mixing and weathering of soil. Roots of the higher plants take active part in the disintegration of rocky mass and also make the compact soil loose. Many rodents, insects and earthworms turn over the soil and sometimes also expose the rock surface for physical and chemical weathering. Burrowing animals, such as rodents, bring soil from deeper regions to the surface. The excreta of soil animal is deposited on the surface of soil in the form of casts which increases the fertility of the soil.
6. Soil aeration improvement. Soil micro-organisms improve aeration of soil. Burrowing worms are also helpful in improving the aeration and percolation.
7. Improvement in aggregation of soil particles. Bacteria, blue-green algae, and some other micro-organisms secrete mucilaginous substances which bind the soil particles into soil aggregates.
8. Injury to plants. Not all the soil organisms are beneficial in their properties and behaviour. Some microbes become parasites of higher plants and cause considerable damage. Nematodes are important animals which cause a number of diseases in plants. Besides these, many soil bacteria and fungi cause many diseases, such as damping off, seedling blight, root rot, mildew diseases in a number of crops.

Check Your Progress

21. What are the components of soil?
22. List the factors responsible for the determining the density of microbial population in the soil.
23. Give Example of some microbes known for nitrogen fixation.

1.7 ANSWERS TO ‘CHECK YOUR PROGRESS’

1. Chemo-organotrophy is the type of metabolism where energy comes from organic chemicals, i.e., compounds containing carbon.
2. Chemolithotrophy refers to the type of metabolism where energy comes from inorganic chemicals (chemical compounds lacking carbon).
3. A few organisms can switch between heterotrophy mode of nutrition when organic carbon is available and autotrophic mode of nutrition when food sources run out; these organisms are referred to as mixotrophs
4. Examples of secondary metabolites are pigments, alkaloids, drugs, essential oils, antibiotics, ergot alkaloids, nucleosides, quinolines, peptides, phenazines, naphthalenes, terpenoids, lectins, polymeric substances and lectins.
5. An extreme environment is a habitat which is characterized by harsh environmental conditions i.e., conditions above or below the optimal range necessary for the growth, development as well as reproduction of the organism.
6. Xeric environments are arid habitats having limited water supply. Cold and hot deserts are few instances of these extreme environments. Example of xeric environments are Sahara Desert.
7. Extremophiles are living organisms that have the ability to survive and thrive well in extreme environments.
8. The optimum temperature required for growth of psychrophile is around 15°C.
9. Osmophiles refers to a group of organisms that are capable to survive in environments with high osmotic pressures.
10. Aquatic microorganisms include members of the bacteria, protozoa as well as fungi.
11. Many microorganisms play foundational roles in aquatic ecosystems, capturing the sun's energy through photosynthesis and, through their role in decomposition, releasing nutrients stored in organic tissue.
12. Microbial photo-synthesizers include algae and cyanobacteria are considered as primary producers in the aquatic system.
13. Eutrophication refers to the process in which a water body becomes overly enriched with nutrients, leading to excessive growth of growth of algae and plankton.
14. Biological oxygen demand (BOD) or Biochemical Oxygen demand (BOD) is referred as the amount of oxygen needed or demanded by aerobic microorganisms to break down the organic matter present in a certain sample of water at a specific temperature over a given period of time.
15. Symbiosis refers to as an evolved interaction or close living relationship between organisms from different species, usually with benefits to one or both of the individuals involved.

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16. Service-service interaction is one of the rarest forms of symbiotic interactions. As the name suggests, both the interacting species receive a service, like shelter or protection from predators.
17. Commensalism refers to a symbiotic association where one species is benefitted from, and is often entirely dependent on the other for resources like food, shelter, or locomotion, with no noticeable adverse or beneficial effects on the host.
18. An endosymbiont is any organism that lives either in specialized structures or inside any body part of another organism.
19. The roots of most species of seed plants (at least 80%) have a symbiotic relationship with soil fungi in which both organisms benefit. The associations are known as mycorrhizae.
20. Mycorrhizae are beneficial to their host plants by secreting hormones or antibiotic agents that reduce the potential of plant disease.
21. The soil is made up of the following components:
 - (1) Mineral particles,
 - (2) Dead organic matter or humus,
 - (3) Soil atmosphere;
 - (4) Soil water, and
 - (5) Biological system or soil micro-organisms.
22. Density of microbial population is actually governed and influenced by climatic conditions, physical and chemical nature of soil and vegetation cover.
23. Azotobacter, Clostridium pasteurianum, actinomycetes, fungi, purple bacteria and a number of blue-green algae are known to fix free atmospheric nitrogen gas into nitrogenous compounds, such as nitrates and nitrites,

1.8 SUMMARY

- Microorganisms are extremely widespread, however, within the microbial world there is also a remarkable number of different metabolic pathways.
- Chemo-organotrophy is the type of metabolism where energy comes from organic chemicals, i.e., compounds containing carbon.
- All the living cells requires a lot of carbon, which is part of all proteins, nucleic acids, as well as cellular structures.
- Primary metabolites are highly useful in metabolic process of organisms as some act as a substrate for these processes, while others act as catalysts.
- Secondary metabolites refer to the organic compounds which are not directly involved in the process of growth, development as well as reproduction of the organism.
- Microbes are used widely to generate new molecules of potential medical and therapeutic importance.
- On the basis of pH extreme environments can also be classified as acidic or alkaline.

- Xeric environments are arid habitats having limited water supply.
- Extremophiles include members of all three domains of life; bacteria, archaea, and eukarya.
- Acidophilic microorganisms survive well in extremely low pH natural or artificial environments like acidic lakes, hydrothermal systems, acid sulfate soils, sulfidic regoliths, ores, etc.
- Alkaliphiles refers to a group of extremophiles that can survive and in environments having extremely high pH value.
- Majority of the organisms growing under high alkaline conditions are described to be prokaryotes.
- Xerophile refers to a group of organisms that are capable of surviving and flourishing well in conditions having low availability of water or low water activity.
- Xerophiles are very closely related to halophiles as halophilic environments tend to have low water activity.
- Halophiles refers to a group of extremophiles that require high salt concentrations for their growth as well as survival.
- Barophiles refers to a group of organisms that are capable of growing and flourishing optimally at pressures greater than atmospheric pressure.
- Rock porosity provides interstitial spaces for microbial colonization as well as translucence enables photosynthesis to take place.
- A variety of minerals like quartz are found in soil and rocks which supports different forms of life.
- Metallotolerant refers to a group of microorganisms that are capable of tolerating as well as detoxifying high levels of dissolved heavy metals.
- Radiophiles refers to a group of extremophiles that are capable of surviving the extreme forms of radiations like ionizing radiation (gamma rays) as well as UV radiation.
- Extremophilic enzymes serve as a model systems to study as well to conduct research studies on various aspects of enzymes such as enzyme evolution, enzyme stability, activity, mechanism, protein structure, function, and biocatalyst under extreme conditions.
- Alkaline active proteases, amylases, cellulases, mannanases, lipases, etc. are used in the formulation of heavy-duty laundry as well as dishwashing detergents.
- The enzymes derived from extremophiles can also be used effectively in several bioremediation processes such as removing metallic waste from sewages and industries.
- Symbiosis refers to as an evolved interaction or close living relationship between organisms from different species, usually with benefits to one or both of the individuals involved.
- The symbiotic association that exist between whales and barnacles is one of the well-known example of commensalism.

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- As learnt from the different kind of interactions, symbiosis is the condition in which two different species stays together and exhibit spatial and temporal overlap in their interactions.
- A few sponges and beetles defend themselves against predators by making use of poisonous polyketide compounds produced by symbiotic bacteria.
- Corals are made up of animals known as corals polyps.
- *Trichonympha* is a genus of single-celled, anaerobic parabasalids of the order *Hypermastigia*.
- An endosymbiont is any organism that lives either in specialized structures or inside any body part of another organism.
- Sap-sucking insects Mealybugs belongs to the family *Pseudococcidae*. They are referred to as Mealybugs because they are usually covered with a mealy or cotton wax secretion.
- Sharpshooters are members of the suborder *Auchenorrhyncha* which feeds on xylem.
- The dietary contribution of gut microbiota in insect host includes production of essential amino acids, vitamins and release of digestive enzymes.
- The indigenous gut microbiota of an insect provides an effective resistance against colonization by pathogenic bacteria or fungal communities.
- Organisms present in the soils are called soil organisms.
- The mineral constituents of the soil are derived from the parental rocks or regolith.
- Humus is not soluble in water. It is present in soil in the form of organic colloids.
- Algae are found in the top layer of soil under the conditions of constant shade and moisture.
- It is estimated that in soil microflora bacteria form about 90 per cent of the total microbe population.
- Soil organisms take part in a number of processes in the soils.
- In the process of decomposition, complex organic matters are converted into simple organic compounds.
- Many soil organisms including soil fungi and bacteria produce growth stimulating substances such as 3-Indol acetic acid, Gibberellins and Gibberellic acid in the soil.
- Many organisms by their mechanical activities help in mixing and weathering of soil.
- Bacteria, blue-green algae, and some other micro-organisms secrete mucilaginous substances which bind the soil particles into soil aggregates.

1.9 KEY TERMS

- **Chemo-organotrophy:** Chemo-organotrophy is the type of metabolism where energy comes from organic chemicals, i.e., compounds containing carbon.
- **Mixotrophs:** A few organisms can switch between heterotrophy mode of nutrition when organic carbon is available and autotrophic mode of nutrition when food sources run out; these organisms are referred to as mixotrophs.
- **Secondary metabolites:** Secondary metabolites refer to the organic compounds which are not directly involved in the process of growth, development as well as reproduction of the organism.
- **Extreme Environment:** An extreme environment is a habitat which is characterized by harsh environmental conditions i.e., conditions above or below the optimal range necessary for the growth, development as well as reproduction of the organism.
- **Xeric environments:** are arid habitats having limited water supply. Cold and hot deserts are few instances of these extreme environments. Example of xeric environments are Sahara Desert.
- **Psychrophiles:** Psychrophiles are the organism adapted to cold conditions.
- **Thermophiles:** Thermophiles (literally heat lovers) are organisms that grow at temperatures above those (25–40°C) that sustain most life forms. Typically, a thermophile shows maximum growth rates at temperatures above 45°C.
- **Hyperthermophiles:** Hyperthermophiles refers to the organisms that can survive and grow at extremely high temperatures above 80°C.
- **Natural flora:** Microorganisms natural to the water body.
- **Transient flora:** Microorganisms entering the water body from outside environment like from soil, air and through pollutants.
- **Eutrophication:** Eutrophication refers to the process in which a water body becomes overly enriched with nutrients, leading to excessive growth of growth of algae and plankton.
- **Dissolved Oxygen(DO):** DO is the abbreviation used for Dissolved Oxygen. Dissolved oxygen characterizes the concentration of oxygen in an aqueous solution and refers to the free oxygen dissolved in water.
- **Turbidity:** Turbidity refers to the measure of relative clarity of a liquid. It is an optical characteristic of water and is a measurement of the amount of light that is scattered by material present in the water when a light is passed through the water sample.
- **Nitrification:** It is the process by which the ammonia is converted to nitrites and then to nitrates by nitrifying bacteria.
- **Symbiosis:** Symbiosis refers to as an evolved interaction or close living relationship between organisms from different species, usually with benefits to one or both of the individuals involved.

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- **Mutualisms:** Mutualisms is a form of symbiosis wherein both the interacting species benefit from the interaction.
- **Commensalism:** Commensalism refers to a symbiotic association where one species is benefitted from, and is often entirely dependent on the other for resources like food, shelter, or locomotion, with no noticeable adverse or beneficial effects on the host.
- **Mycorrhizae:** The roots of most species of seed plants (at least 80%) have a symbiotic relationship with soil fungi in which both organisms benefit. The associations are known as mycorrhizae.
- **Ericoid mycorrhizas:** These occur on plants which have very fine, hair-like roots and cortex is composed of only a few cells.
- **Soil organisms:** Organisms present in the soils are called soil organisms.
- **Decomposition:** In the process of decomposition, complex organic matters are converted into simple organic compounds.

1.10 SELF-ASSESSMENT QUESTIONS AND EXERCISES

Short-Answer Questions

1. Write the four broad categories of metabolic diversity of microbes.
2. State the difference between chemo-organotrophy, chemolithotrophy and phototrophy?
3. How knowledge of microbial metabolism has been useful in the advancements of science?
4. How the extreme environments can be classified on the basis of extreme physiochemical conditions?
5. Which organisms are known as extremophiles?
6. Write the difference between thermophiles and hyperthermophiles.
7. Differentiate between natural flora and transient flora.
8. How does microbes act as decomposer of aquatic system?
9. List the factors influence the BOD of water.
10. What is turbidity?
11. How does microbes fix atmospheric nitrogen into useable form and increase the fertility of soil?
12. In what ways soil micro-biome can contribute to the health of the crop?
13. What are the two types of symbioses interactions?
14. Which organisms are called endosymbionts?
15. What is bioluminescence?

Long-Answer Questions

1. Explain a few characteristic features of primary metabolites.
2. Describe the application and potential use of extremophiles in biotechnology.
3. Analyze the role of microbes in aquatic environment.
4. Elaborate on the common methods of biological treatment of waste.
5. Explain the role of microbes in recycling of soil nutrients.
6. Describe the types of symbiotic relationship in detail.
7. Elaborate on the dietary contribution of gut microbiota in an insect host.
8. Describe the four principal types of mycorrhizas based on morphological and anatomical features.
9. Analyze the flora and fauna of the soil.
10. Write an explanatory note on the role of soil organisms in formation of soil.

NOTES

1.11 FURTHER READING

Atlas, Ronald M., Richard Bartha. 1993. *Microbial Ecology Fundamentals and Applications*. U.S.: Benjamin/Cummings Publishing Company.

Chénard, Caroline, Federico M. Lauro. 2017. *Microbial Ecology of Extreme Environments*. U.S.: Springer International Publishing.

Alexander, Martin. 1991. *Introduction to Soil Microbiology*. U.S.: R.E. Krieger Publishing Company

David, L. Kirchman. 2012. *Processes in microbial ecology*. Oxford: Oxford University Press.

UNIT 2 MICROBIAL ECOLOGY II

Structure

- 2.0 Introduction
- 2.1 Objectives
- 2.2 Role of Microbes in Biogeochemical and Nutrient Cycling Process
 - 2.2.1 Role of Microbes in Carbon Cycle
 - 2.2.2 Role of Microbes in Nitrogen Cycle
 - 2.2.3 Role of Microbes in Phosphorous Cycle
 - 2.2.4 Role of Microbes in Sulphur Cycle
- 2.3 Role of Microbes in Metal Cycle
 - 2.3.1 Role of Microbes in Silicon Cycle
 - 2.3.2 Role of Microbes in Iron Cycle
 - 2.3.3 Role of Microbes in Manganese Cycle
- 2.4 Aquatic Micro - Organisms
- 2.5 Role of Microorganism in Water Purity Test
- 2.6 Sewage Waste Water Treatment
- 2.7 Answers to 'Check Your Progress'
- 2.8 Summary
- 2.9 Key Terms
- 2.10 Self-Assessment Questions and Exercises
- 2.11 Further Reading

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2.0 INTRODUCTION

Microorganisms play a vital role in every ecological community by serving both as producers and as decomposers. Although plants are the most common primary producers, autotrophic photosynthetic microbes (such as cyanobacteria and algae) can harness light energy to generate organic matter. Additionally, in zones where light cannot penetrate (and thus photosynthesis cannot be the basic means to produce energy), chemosynthetic microbes provide energy and carbon to the other organisms in the ecosystem. Other microbes are decomposers, with the ability to recycle nutrients from dead organic matter and other organisms' waste products.

By virtue of their omnipresence, microbes impact the entire biosphere; indeed, microbial metabolic processes (including nitrogen fixation, methane metabolism, and sulfur metabolism) collectively control global biogeochemical cycling. The ability of microbes to contribute substantially to the function of every ecosystem is a reflection their tremendous biological diversity. Microbial life plays a primary role in regulating biogeochemical systems in virtually all of our planet's environments, including some of the most extreme, from frozen environments and acidic lakes, to hydrothermal vents at the bottom of deepest oceans, and some of the most familiar, such as the human small intestine.

We almost always presume microorganisms are harmful to us. So this is because we read about how they cause diseases to both plants and animals including humans. But, it is a fact that microorganisms are useful to us in many ways.

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Microorganisms help in the production of many food items, making medicines, keeping the environment clean, in manufacturing and in research. Biota are an essential component of most sewage treatment processes and many water purification systems.

In this unit you will study about role of microbes in biogeochemical and nutrient cycling processes, microbes in metal cycling, aquatic micro-organisms, fresh water microbiota, sea water microbiota, role of micro-organisms in water purity and water purity test, and sewage waste water treatment.

2.1 OBJECTIVES

After going through this unit you will be able to:

- Analyze the role of microbes in biogeochemical and nutrient cycling processes
- Understand the role of microbes in metal cycling
- Elaborate aquatic micro-organisms
- Explain the role of micro-organisms in water purity and water purity test
- Comprehend sewage waste water treatment

2.2 ROLE OF MICROBES IN BIOGEOCHEMICAL AND NUTRIENT CYCLING PROCESS

A biogeochemical cycle is referred to as the pathway via which a chemical substance is turned over or moves through the biotic (biosphere) and the abiotic (lithosphere, atmosphere and hydrosphere) compartments of Earth. The important biogeochemical cycles are: - calcium cycle, carbon cycle, hydrogen cycle, mercury cycle, nitrogen cycle, oxygen cycle, water cycle, phosphorus cycle, selenium cycle, iron cycle, silica cycle, sulphur cycle as well as rock cycle. A few characteristics of biogeochemical cycles are as follows:

- *Biological Chemical + Geological Process = Biogeochemical*
- Energy flows via an ecosystem and is released as heat, however chemical elements are recycled.
- The ways in which an element or compound moves between its numerous biotic and abiotic forms & locations in the biosphere is referred to as biogeochemical cycle. Precisely, it can be defined as movement of nutrients as well as other elements between living and non-living components of ecosystem.
- Sun is the primary source of energy on earth. Life on earth comprises a great variety of living organisms. These living organisms occur and survive

in a diversity of associations. Such survival encompasses the presence of systemic flows such as flows of energy, water as well as nutrients.

- The balance of the chemical elements is preserved by a cyclic movement through the tissues of plants and animals.
- The cycle begins by absorbing the chemical elements by the organism and is returned to the atmosphere, hydrosphere or lithosphere via decomposition. These cycles are mainly energized by solar insolation.

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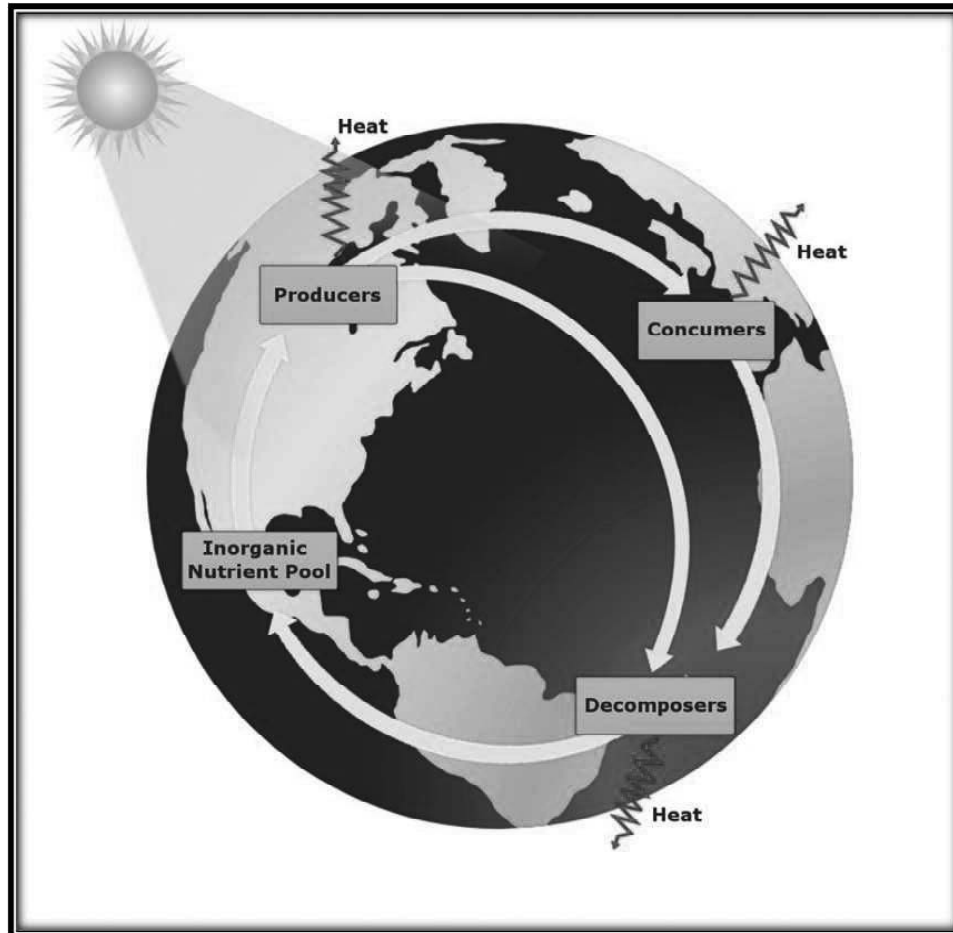


Fig. 2.1 Basic Concept of Biogeochemical Cycle.

Figure 2.1 is depicting the basic concept of biogeochemical cycle.

Types of biogeochemical cycles

There are two types of biogeochemical cycles:

(1) The Gaseous Cycle

In the gaseous cycle, the main reservoir of nutrients is either the atmosphere or the hydrosphere (ocean). For instance: - nitrogen, oxygen, carbon and water cycles are gaseous cycles.

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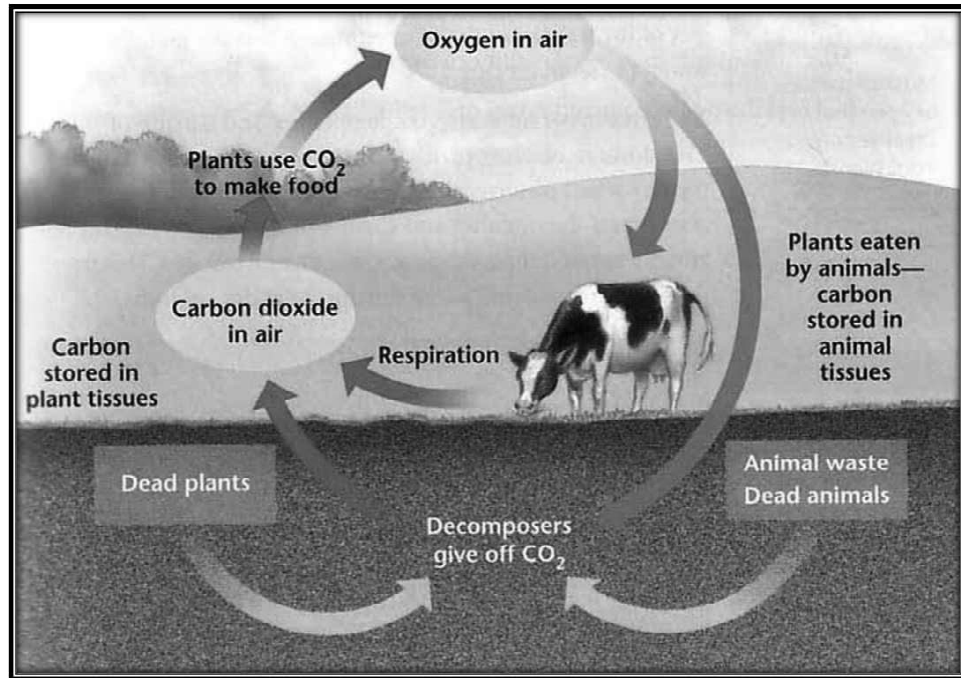


Fig. 2.2 Gaseous Cycle (Oxygen) Operating In Atmosphere

Figure 2.2 is depicting the gaseous cycle (oxygen) operating in atmosphere.

(2) The Sedimentary Cycle

In the sedimentary cycle, the main reservoir is the lithosphere and the sedimentary as well as other rocks of the earth's crust. For instance: - iron, calcium, phosphorus, and other more earthbound elemental cycles are sedimentary cycles.

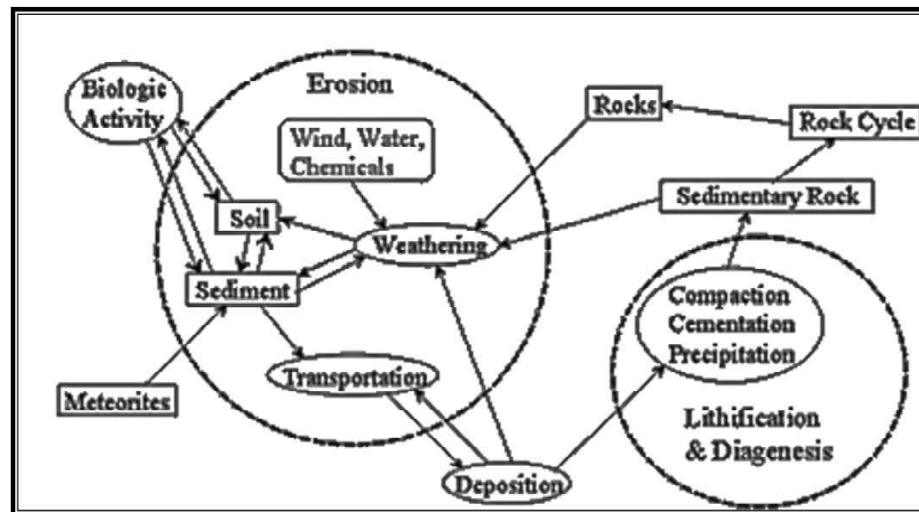


Fig. 2.3 Generalized Sedimentary Cycle

Figure 2.3 is depicting the generalized sedimentary cycle.

Input of Nutrients (Addition) To Ecosystem

Nutrients enter into the ecosystem by four ways:

(a) Rock weathering

Essential nutrients added to the ecosystem by this process are: Calcium (Ca), magnesium (Mg), potassium (K), sodium (Na), silicon (Si), iron (Fe), aluminum (Al), and phosphorus (P).

(b) Atmospheric inputs

Atmosphere adds nutrients to the ecosystem via a number of biological processes like carbon is absorbed by the process of photosynthesis, sodium and potassium deposited via precipitation.

(c) Biological Nitrogen Fixation

This process is majorly for the fixation of nitrogen carried out by number of organisms like several species of bacteria, a few actinomycetes (fungi) & blue-green algae (cyanobacteria).

(d) Immigration

Migration and subsequent death of the organism can add significant nutrients to the ecosystem.

Output of Nutrients (Loss) from ecosystem

- (a) **Erosion:** Soil erosion leads to removal of top most layer of soil rich in essential nutrients.
- (b) **Leaching:** Leaching occurs when water flowing vertically through the soil transports nutrients in solution downward in the soil profile. The process can lead to loss of essential nutrients.
- (c) **Gaseous losses:** Gaseous loss of nutrients can occur when specific environmental conditions promote the export of nutrients in a gaseous form.
- (d) **Emigration and Harvesting:** Emigration of animals and the removal of vegetation can also lead to huge loss of nutrients.

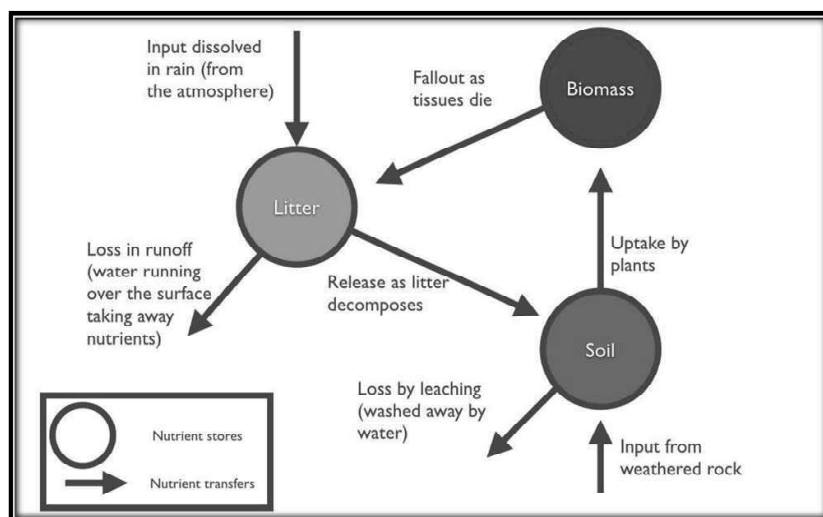


Fig. 2.4 Flow of Nutrients in an Ecosystem

Figure 2.4 is depicting the flow of nutrients in an ecosystem.

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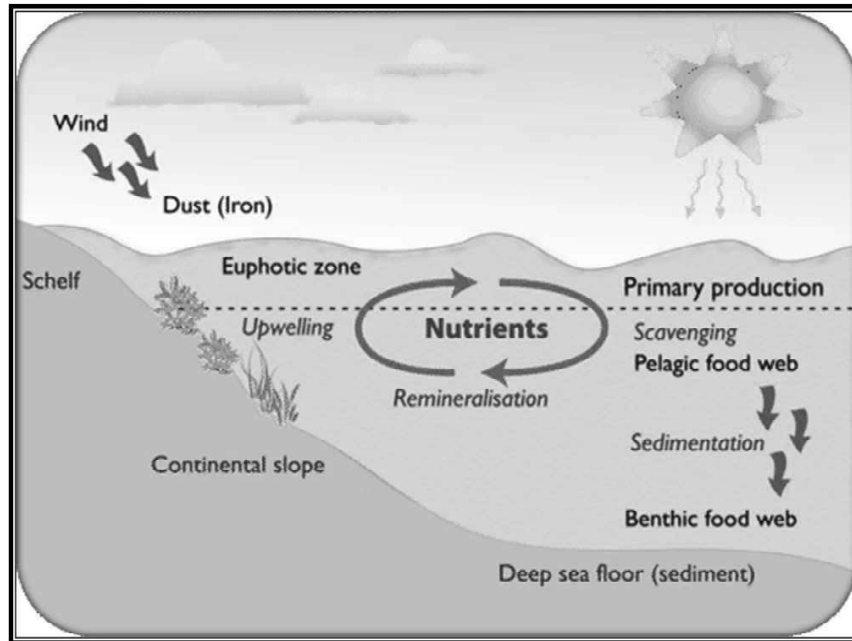


Fig. 2.5 Nutrient Cycle via Aquatic Ecosystem

Figure 2.5 is depicting the nutrient cycle via aquatic ecosystem.

Types of Nutrient Flow in Ecosystem

Two major types of system are as follows:

- (1) **Closed system-** In the Closed system, chemicals or elements used in the ecosystem are recycled instead of being lost.

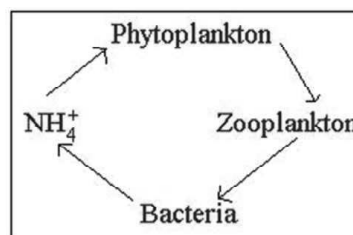


Fig. 2.6 Closed Type of Nutrient Flow in Ecosystem

Figure 2.6 is depicting closed type of nutrient flow in ecosystem.

- (2) **Open system:** In the open system, chemicals or elements tend to flow in and out of the system.

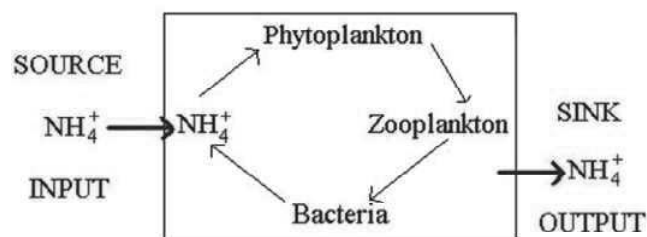


Fig.2.7 Open Type of Nutrient Flow in Ecosystem

Figure 2.7 is depicting open type of nutrient flow in ecosystem.

2.2.1 Role of Microbes in Carbon Cycle

Carbon dioxide is the basic building block that most autotrophs use to build multi-carbon, high-energy compounds like glucose. The energy obtained from the sun is used by autotrophs (like green plants) to form the covalent bonds that link carbon atoms together. These chemical bonds store this energy for later use in the process of respiration. Most of terrestrial autotrophs obtain the carbon dioxide directly from the atmosphere, however, marine autotrophic organism acquire it in the dissolved form (carbonic acid, H_2CO_3). The fixation of carbon dioxide leads to the production of oxygen- a by-product of the process.

Heterotrophs obtain the high-energy carbon compounds by consuming autotrophs as they are not able to produce their own food. Heterotrophs break down the organic compound during the process of respiration to release energy in the form of ATP. The most efficient type of respiration, aerobic respiration, requires oxygen obtained from the atmosphere or dissolved in water. Thus, there is a constant exchange of oxygen and carbon dioxide between the autotrophs (which need the carbon) and the heterotrophs (which need the oxygen). Gas exchange through the atmosphere and water is one way that the carbon cycle connects all living organisms on Earth.

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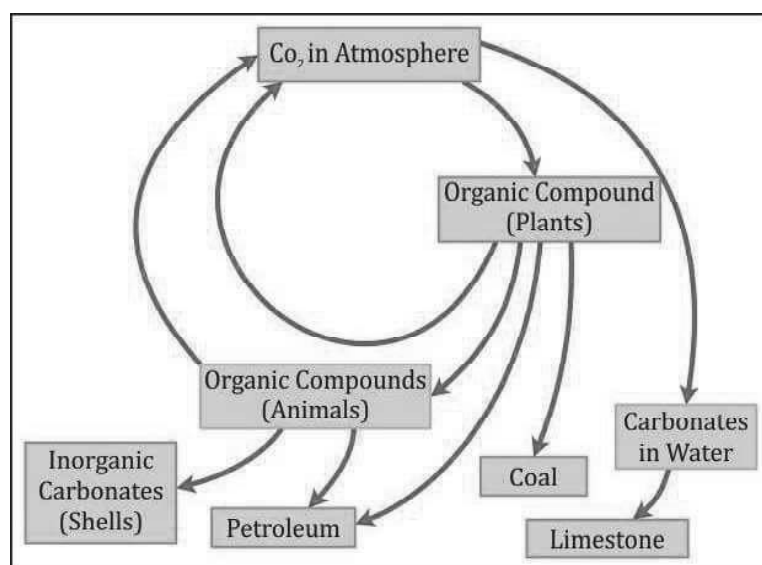


Fig. 2.8 Carbon Cycle

Figure 2.8 depicting the carbon cycle.

- Carbon is basically a minor constituent of the atmosphere as compared to oxygen as well as nitrogen.
- However, life is not possible on earth surface without carbon dioxide
- It is vital for the production of carbohydrates through photosynthesis by plants.
- Carbon dioxide is the element which anchors all organic substances from coal and oil to DNA (deoxyribonucleic acid: the compound that carries genetic information).

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- Carbon is present in the atmosphere, mainly in the form of carbon dioxide (CO₂).
- Carbon cycle involves continuous exchange of carbon between the atmosphere as well as organisms.
- Carbon from the atmosphere moves to green plants via the process of photosynthesis (it refers to the process by which plants can convert the inorganic raw material like carbon dioxide and water to organic compounds like glucose in the presence of sunlight), and then to animals.
- By process of respiration and decomposition of dead organic matter, it returns to the atmosphere.
- Carbon-dioxide cycle is usually a short-term cycle. However, some carbon also enters a long-term cycle. It gathers as un-decomposed organic matter in the peaty layers of marshy soil or as insoluble carbonates in bottom sediments of aquatic systems which take a long time to be released.
- In deep oceans, such carbon can remain buried for millions of years till geological movement may lift these rocks above sea level.
- These rocks may be exposed to erosion, releasing their carbon dioxide, carbonates and bicarbonates into streams and rivers.
- Fossil fuels such as coals, oil and natural gas etc. are organic compounds that were buried before they could be decomposed and were subsequently transformed by time and geological processes into fossil fuels. When they are burned the carbon stored in them is released back into the atmosphere as carbon dioxide.

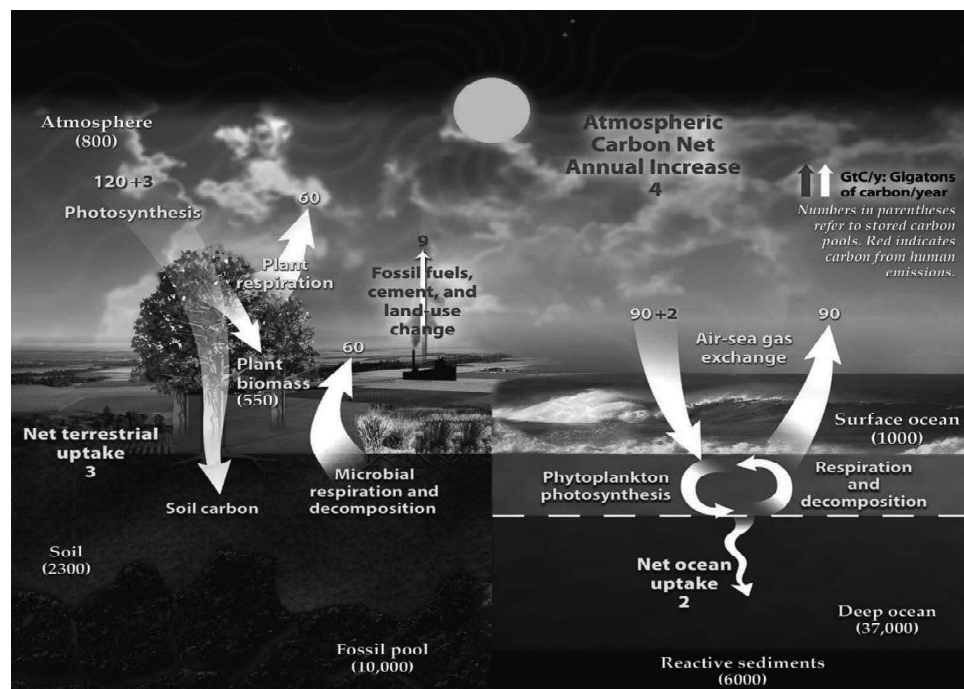


Fig. 2.9 Carbon Cycle

Figure 2.9 is depicting the carbon-cycle.

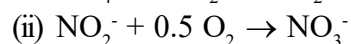
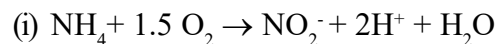
2.2.2 Role of Microbes in Nitrogen Cycle

Nitrogen is vital for several processes and is crucial for life on Earth. The nitrogen cycle is the biogeochemical cycle by which nitrogen is converted into multiple chemical forms as it circulates among atmosphere, hydrosphere and lithosphere. Nitrogen cycling in aquatic Ecosystem can be divided into five phases

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(1) **Nitrogen fixation-** Nitrogen fixation refers to the conversion of N_2 to ammonia or related nitrogenous compounds by Blue green algae (Cyanobacteria) like Anabaena, Nostoc, Azolla, etc., In the process, nitrogen undergoes a set of changes, in which two nitrogen atoms get separated and combine with hydrogen to form ammonia (NH_4^+).

(2) **Nitrification-** Nitrifying bacteria known as chemoautotrophs are responsible for carrying out the process of nitrification. The term nitrification refers to the conversion of ammonium to nitrate. First step is carried out by Nitrosomonas sp whereas second step is carried out by Nitrobacter sp. Or Nitrospira sp. Both steps/reactions use NH_4^+ and NO_2^- as an energy source whereas CO_2 as a carbon source. This is a non-photosynthetic type of growth. NH_3 and NH_4^+ are both either assimilated by aquatic plants for growth or nitrified (oxidized) to NO_3^- (nitrate). Nitrate can also be used as a growth substrate. It is a two-step process:



These oxygen-driven reactions works best at pH 7-8 and at a temperature of 25-30 degree Celsius. However, under low dissolved oxygen conditions, it runs in reverse direction. It occurs in the hypolimnion under eutrophic (stagnant) conditions.

(3) **Assimilation-** Plants take in the nitrogen compounds which are available in the form of ammonia, nitrite ions, nitrate ions or ammonium ions and are used in the formation of the plant as well as animal proteins. By this way, it enters the food web when the primary consumers eat the plants.

(4) **Ammonification-** When plants or animals die, the nitrogen present in the organic matter is released back into the soil. The decomposers, namely bacteria or fungi present in the soil, convert the organic matter back into ammonium. This process of decomposition produces ammonia, which is further used for other biological processes.

(5) **Denitrification** - By this process, NO_3^- in water is converted into atmospheric N_2 , nitric oxide or nitrous oxide. This must occur under anaerobic conditions (anaerobic respiration). The process is carried out by bacteria like *Pseudomonas sp.*, *Alkaligenes sp.* and *Bacillus sp.*

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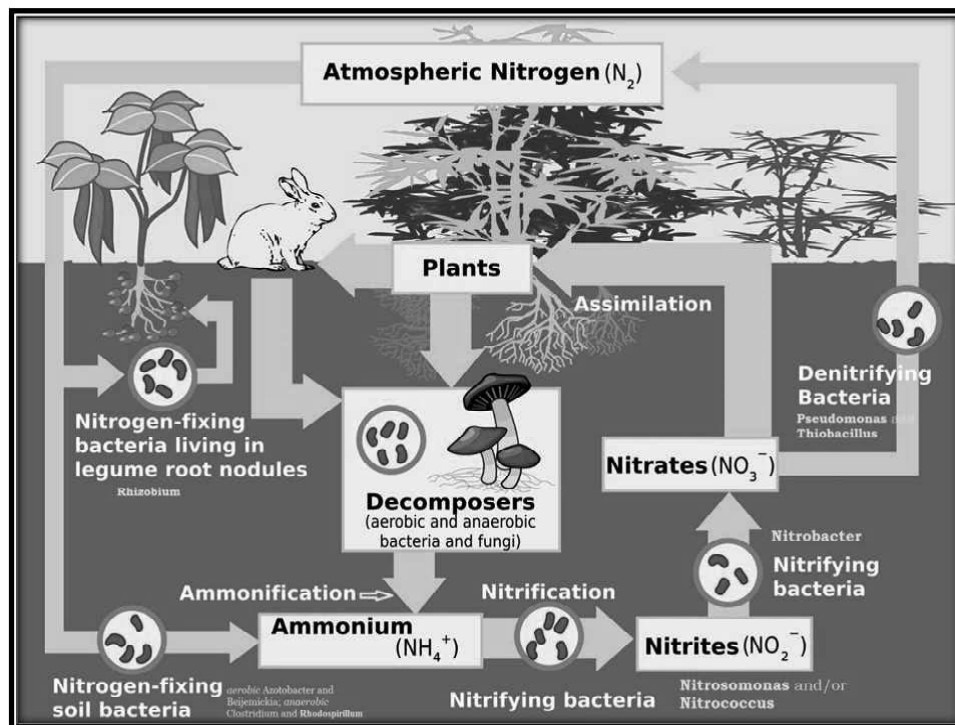


Fig. 2.10 Nitrogen Cycle

Figure 2.10 is depicting the nitrogen cycle.

2.2.3 Role of Microbes in Phosphorous Cycle

Phosphorous is an essential component of biomolecules like ADP and ATP, nucleic acids, phospholipids (membranes), apatite (bones and teeth), etc. In one of the study conducted (oceanographer *Redfield* in the 1950's), Phosphorous is considered to be the most limiting nutrient in the fresh-water system because the amount of phosphorous available to organisms is much less than the amount required when compared to other elements. In freshwater, P (Phosphorous) is often 80,000 X less concentrated than the amount required by phytoplankton.

Forms and Measurement of P

Total Phosphorous (P) = DIP + DOP + PP

- DIP** – (<5%) **Dissolved Inorganic Phosphorus** - PO_4^{3-} polyphosphates
- DOP** – **Dissolved Organic Phosphorus** -often associations of organic colloids. Alkaline phosphatase enzyme facilitates the release of P from these organic compounds
- PP Particulate Phosphorus**-often largest percentage of P in lakes (>70%) – nucleic acids (decompose slowly), phosphate sugars, ATP (available more quickly, most P is in organic matter living or dead organisms, some particulate P is mineral P

Sources of Phosphorous

- **Rock Weathering**- It involves weathering of calcium phosphate minerals, especially apatite [$Ca_5(PO_4)_3OH$]. However, it is a slow process and happens over a period of time.

- **Phosphorous** is mostly stored in marine deep ocean sediments
- **Water Pollution-** All the sources of water pollution can be roughly divided into two categories:-
 - (1) **Point Source Pollution:** Point source pollution refers to contaminants or harmful/toxic compounds that enter into aquatic bodies via a fixed source such as a pipe or ditch. For instance: Discharges pipes from a sewage treatment plant of factories or several other organizations.
 - (2) **Non-Point Pource Pollution:** Non-point source pollution resulting from diffuse sources in contrast to point source pollution. Nonpoint source pollution generally results from land runoff, precipitation, atmospheric deposition, drainage, seepage, or hydrological modification where tracing pollution to a single source becomes extremely difficult.

Modes of Entry of P to aquatic systems.

- (i) **Precipitation** – dust in air
- (ii) **Groundwater** –P adsorbs to soil particles
- (iii) **Surface runoff**

Steps of Phosphorous Cycle

Steps involved in the phosphorous cycle are as follows:

- (1) **Entry of phosphorous into aquatic system:** Phosphorous can enter inside aquatic system via different modes as described in the above section.
- (2) **Absorption by Plants:** The phosphate salts dissolved in water are absorbed by the plants. The aquatic plants absorb inorganic phosphorus from lower layers of water bodies. Since phosphate salts do not dissolve in water properly, they affect plant growth in aquatic ecosystems.
- (3) **Absorption by Animals:** The animals absorb phosphorus from the plants or by consuming plant-eating animals.
- (4) **Return of Phosphorus Back to the Ecosystem:** When the plants and animals die they are decomposed by microorganisms. During this process, the organic form of phosphorus is converted into the inorganic form, which is recycled back to water. Water will end up in sediments or rocks, which will again release phosphorus by weathering. Thus, the phosphorus cycle starts all over again.

Decomposition and Excretion

- (i) Biota persist due to well-developed, efficient recycling of Phosphorous
- (ii) Phosphorous excreted by animals is rapidly taken up by algae as well as bacteria present in aquatic system.
- (iii) Decomposition helps in the liberation of usable P
- (iv) $\text{iv. organic } - P \rightarrow \text{PO}_4^{3-}$

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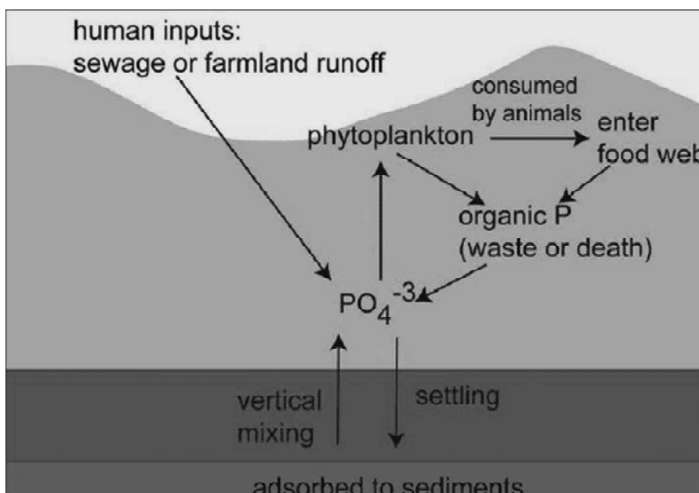


Fig. 2.11 Phosphorous Cycle

Figure 2.11 is depicting the Phosphorous cycle in aquatic system.

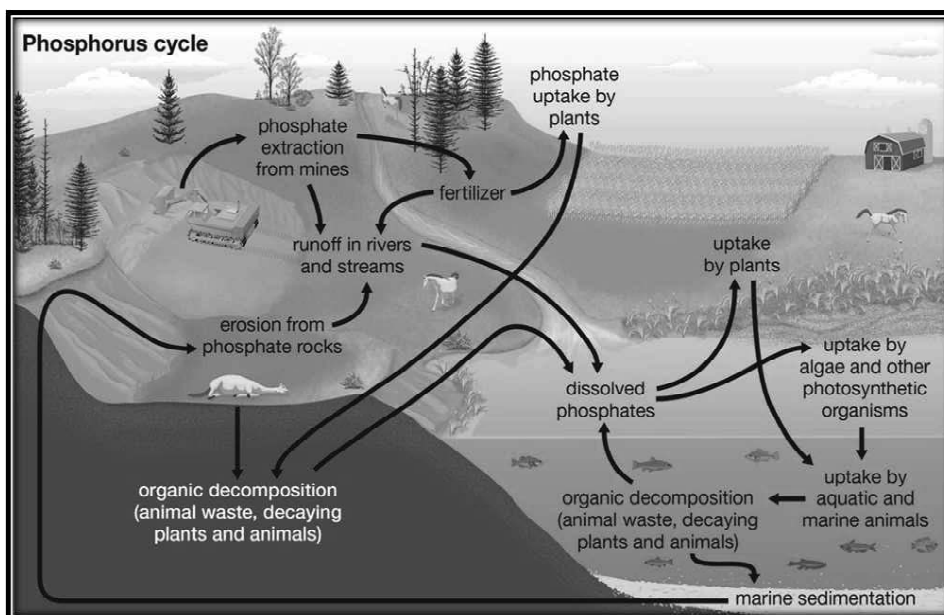


Fig. 2.12 Phosphorous Cycle

Figure 2.12 is depicting the Phosphorous cycle.

2.2.4 Role of Microbes in Sulphur Cycle

Sulphur is a yellow, brittle, tasteless, odourless non-metal. It is one of the most abundant elements in nature and is present in almost all kinds of proteins. Sulphur is released into the atmosphere via:

- Burning of fossil fuels
- Volcanic activities
- Decomposition of organic molecules.

On land, sulphur is stored in underground rocks and minerals. It is released by:

- Precipitation
- Weathering of rocks
- Geothermal vents.

There occurs a continuous loss of sulphur from terrestrial ecosystems as some of it drains into lakes as well as streams and eventually into the ocean as runoff. There are several natural sources like volcanic eruptions, evaporation of water, and breakdown of organic matter in swamps, that release sulphur directly into the atmosphere. This sulphur falls on earth with rainfall.

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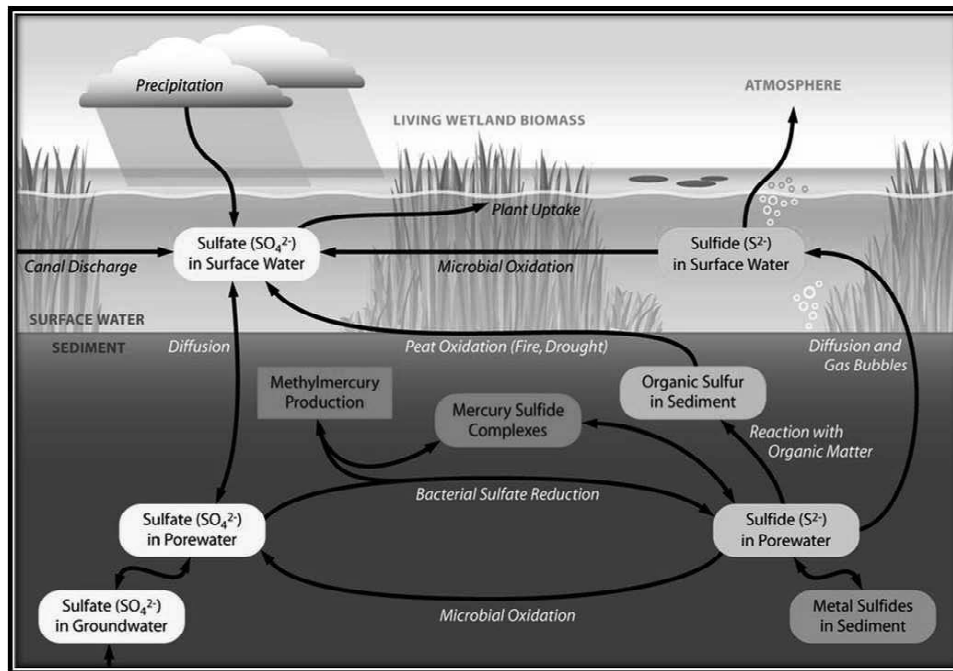


Fig. 2.13 Sulphur Cycle

Figure 2.13 is depicting the sulphur cycle.

Sulphur Cycle

The sulphur cycle is described below:

- The sulphur is released by geochemical and meteorologic processes like the weathering of rocks.
- When the released Sulphur comes in contact with air, it gets converted into sulphates (SO_4).
- These Sulphates are taken up by plants as well as microorganisms and are converted into organic forms.
- Animals acquire these organic forms of sulfur from their foods.
- Death and decomposition of the animals leads to release of some of the sulphur while some enter the tissues of microbes.

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Steps of Sulphur Cycle

Sulfur cycling, one of the active biological processes in aquatic sediments, is catalyzed by sulfate-reducing prokaryotes (SRPs) and sulfur-oxidizing prokaryotes (SOPs).

Following are the important steps of the sulphur cycle:

(1) Decomposition of Organic Compounds

Sulfate is one of the major ions in lake waters. Under anaerobic conditions, in which bacteria persist in the oxidation of biological material, hydrogen sulfide is produced. Further, hydrogen sulfide occurs, when anoxic situations happen in the deep waters just above the sediments, and the water is sufficiently acidic to precipitate the iron present.

(2) Oxidation of Hydrogen Sulphide to Elemental Sulphur

Hydrogen sulphide oxidises to produce elemental sulphur. Certain photosynthetic bacteria from the families Chlorobiaceae and Chromatiaceae initiate the oxidation process.

(3) Oxidation of Elemental Sulphur

Elemental sulphur is converted into sulphates by chemolithotrophic bacteria.

(4) Reduction of Sulphates

Sulphates are reduced to hydrogen sulphide by *Desulfovibrio desulfuricans*. This occurs in two steps:

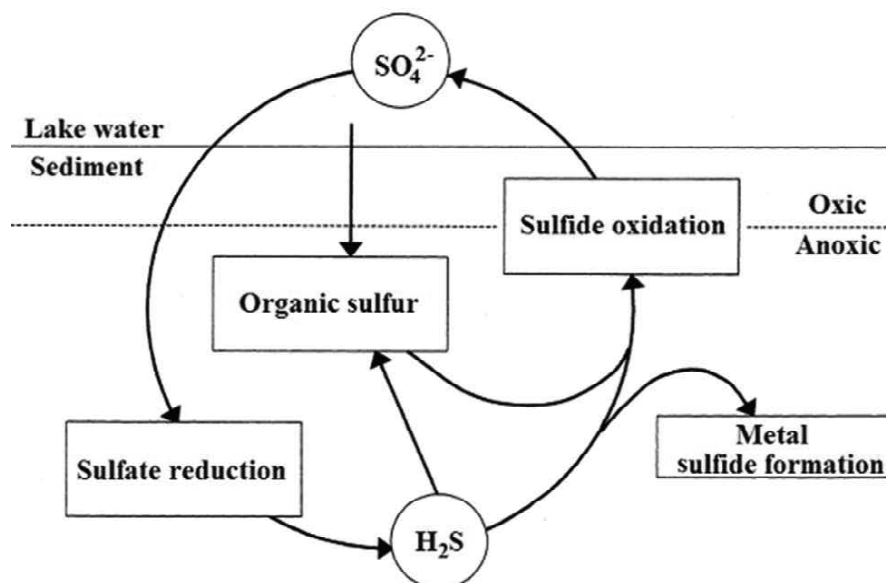
- Firstly, the sulphates are converted to sulphites utilizing ATP.
- Secondly, the reduction of sulphite to hydrogen sulphide.

Once in the aquatic system, some of the sulphur cycles via freshwater communities as it moves through food chains, some re-enters the atmosphere, and some is lost to the freshwater depths where it combines with iron to form ferrous sulfide (FeS). Sulfur re-enters the atmosphere as:

- (a) **Anaerobic respiration** by sulfate-reducing bacteria causes the release of hydrogen sulfide (H₂S) gas especially from marshes, tidal flats, and similar environments in which anaerobic microorganisms thrive
- (b) **Volcanic activity** releases additional but much smaller amounts of sulfur gas into the atmosphere.

Anthropogenic activities have contributed largely to the amount of sulphur present in ecosystem. Rapid burning of fossil fuels due to industrialization as well as over use of vehicles has contributed much to the acidification of ecosystems. The major contributors to freshwater acidification are sulphur oxides and nitrogen oxides. Increases in concentration of sulphate in runoff (due to increased acidity inputs) is associated with an increase in base cation run-off as well as bicarbonate decrease, producing the acidifying effect as observed in freshwater ecosystems. In a natural state, most of the nitrogen entered into freshwater ecosystems will be used by vegetation. However, when present in excess amounts, all of the nitrogen is unable to be utilized by vegetation, and excess nitrogen is found as nitrate in the

water's runoff. Nitrate as well as sulfate contribute to the acidification of the aquatic system in the same way.



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Fig. 2.14 Sulphur Cycle

Figure 2.14 is depicting the sulphur cycle in freshwater system.

2.3 ROLE OF MICROBES IN METAL CYCLE

Microbes play a huge role in the cycling of metals like iron, silicon and manganese on earth's surface. In this section, we shall discuss the role of microbes in the cycling of these metals.

2.3.1 Role of Microbes in Silicon Cycle

Silicon is known to be the second most abundant element in the Earth's crust, 27% by weight (Faure, 1986), exceeded only by oxygen. In natural environments, silicon is most stable in various forms of solid silicon dioxide (silica, SiO_2), as mineral silicates and as well as the dissociated anions of silicic acid (H_4SiO_4). Silica forms the skeleton structures of numerous aquatic plankton like diatoms, radiolarians as well as the spicules of sponges as studied in the previous units. Hence, silica controls primary production in aquatic systems as well as its cycling is controlled by the interplay between biological and physiochemical processes. Silica enters into the aquatic environments by weathering of rock-forming silicate minerals by acidic dissolution. Biological uptake of silica by organisms depletes the surface waters with respect to silica. When these planktonic organisms die, their silica skeletons sink and starts dissolving.

Diatoms are present extensively in freshwater system. They are enclosed in skeleton made up of silica having two perforated valves. The dissolution of biogenic silica in aquatic systems happens when diatom skeleton or skeleton composed of silica is exposed to undersaturated water. The rate of dissolution depends upon aquatic factors like temperature, pH etc. Further, several microbial species can

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significantly increase the rate of dissolution of biogenic silica by releasing enzymes. The destruction, degradation as well as solubilization of mineral silicates is highly influenced by microbial activity. Biofilms of free-living microbes like algae, lichens, fungi, prokaryotes living on the or withing prokaryotes, fungi, and algae, as well as lichens, living on and within the influence the rate as well as mechanism of physical & chemical weathering of siliceous rocks. The amount of dissolved silica present in aquatic environment controls the composition & abundance of planktonic diatoms.

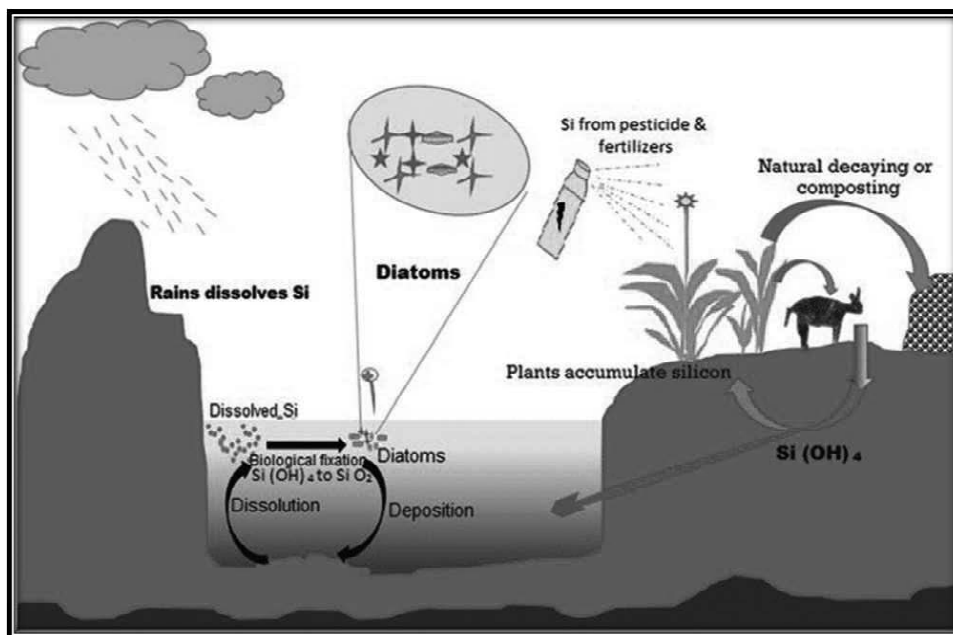


Fig. 2.15 Silicon Cycle

Figure 2.15 is depicting the silicon cycle.

2.3.2 Role of Microbes in Iron Cycle

Iron is an essential micronutrient for almost every life form present on earth. It is a crucial constituent of hemoglobin, vital to nitrogen fixation as part of the Nitrogenase enzyme family as well as part of the iron-sulfur core of ferredoxin it facilitates the process of electron transport in chloroplasts, eukaryotic mitochondria and bacteria. Due to the high reactivity of Fe^{2+} with oxygen and low solubility of Fe^{3+} , iron is a limiting nutrient in aquatic environment and affects the growth of phytoplankton. The iron cycle (Fe) is the biogeochemical cycle of iron via the atmosphere, hydrosphere, biosphere and lithosphere. Iron is suspended as sediment particles in rivers. Generally, iron is available as an inorganic source to phytoplankton; however, organic forms of iron can also be utilized by specific diatoms. Uptake of iron by phytoplankton lowers down iron concentrations in surface seawater. Remineralization, happens when the sinking phytoplankton are degraded by zooplankton as well as bacteria. Iron chiefly is present in particulate phases as ferric iron, and the dissolved iron fraction is removed out of the water column by coagulation.

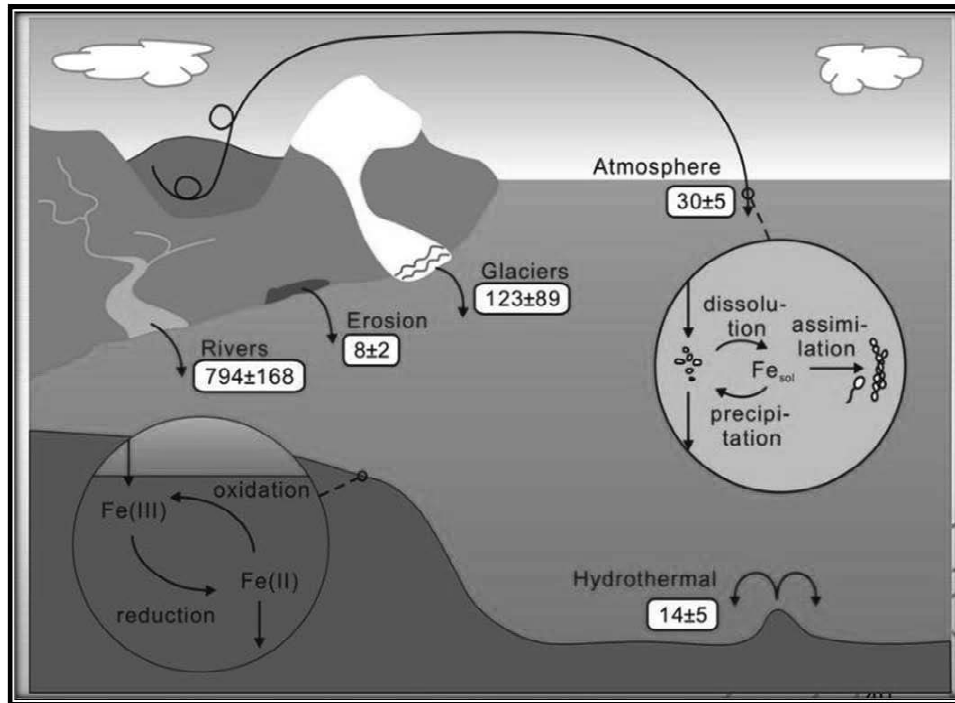


Fig 2.16 Iron Cycle

Figure 2.16 is depicting the Iron cycle.

The iron cycle influences other cycles like nitrogen, phosphorous as well as sulphur cycles. Soluble Fe₂ can act as the electron donor, reducing oxidized organic as well as inorganic electron receptors, including O₂ and NO₃, and become oxidized to Fe₃. The oxidized form of iron can then act as the electron acceptor for reduced sulphur, H₂, and organic carbon compounds. This returns the iron to the oxidized Fe₂ state, completing the cycle. This transition of iron between Fe₂ and Fe₃ inside aquatic environments influences the freshwater phosphorus cycle. Fe₂ gets oxidized to Fe₃ in the presence of oxygen (present in aquatic system). The reaction can be achieved either abiotically or biotically, i.e., by microbes via lithotrophic oxidation. Fe₃ can form iron hydroxides, which has the potential to bind tightly to phosphorus, removing it from the bioavailable phosphorus pool, and hence limiting the primary productivity of the aquatic system. Under anoxic conditions, Fe₃ can be reduced, used by microbes to be the final electron acceptor from either organic carbon or H₂. This leads to the releases of phosphorus back into the aquatic system for biological use. Iron influences sulphur cycle too. For instance: Green and purple sulphur bacteria can utilize Fe₂ as an electron donor during anoxygenic photosynthesis. Sulphate reducing bacteria under anoxic environments can reduce sulphate to sulphide, which then binds to Fe₂ to form iron sulphide. Iron sulphide is a solid mineral that can easily precipitates out of water and removes the iron as well as sulphur. Sulphide can reduce Fe₃ from iron that is previously bound to phosphate when there are no additional metal ions accessible, which releases the phosphate leading to the formation of iron sulphide. Iron plays a significant role in the nitrogen cycle, apart from its role as part of the essential enzymes involved in the process of nitrogen fixation. Under anoxic conditions, Fe₂ can donate an electron that is accepted by NO₃⁻ which is oxidized to several different

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forms of nitrogen compounds, NO_2^- , N_2O , N_2 , and NH_4^+ , while Fe_2 is reduced to Fe_3

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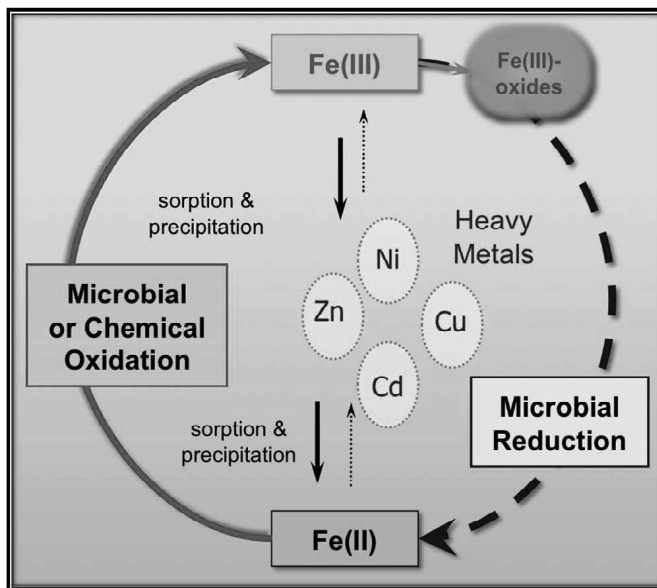


Fig. 2.17 Oxidation of Iron

Figure 2.17 is depicting the oxidation of iron.

2.3.3 Role of Microbes in Manganese Cycle

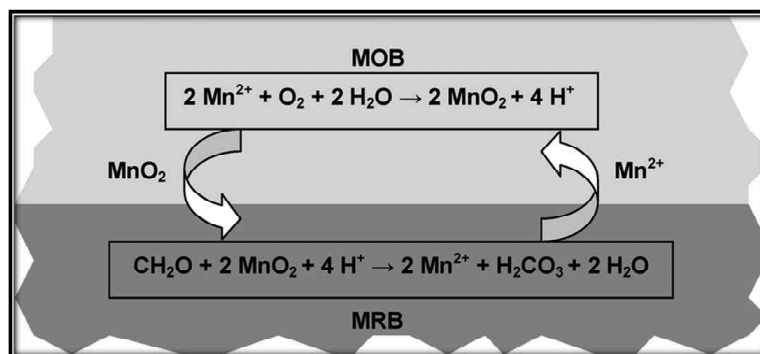


Fig. 2.18 The Reactions of Manganese Cycle

Figure 2.18 is depicting the reaction of manganese cycle in freshwater system.

The manganese cycle is the biogeochemical cycle of manganese via the atmosphere, hydrosphere, biosphere and lithosphere. There are bacteria that oxidise manganese to insoluble oxides, and others that reduce it to Mn^{2+} in order to use it. Manganese cycle occurring in a sediment-water system. The manganese oxidation is performed by Manganese Oxidizing Bacteria (MOB) in an oxic zone, while the manganese reduction is performed by Manganese Reducing Bacteria (MRB)-with concomitant oxidation of organic matter-in an anoxic zone.

Source: Minireview: The Potential of Enhanced Manganese Redox Cycling for Sediment Oxidation Liesje De Schampelaire, Korneel Rabaey, Nico Boon,

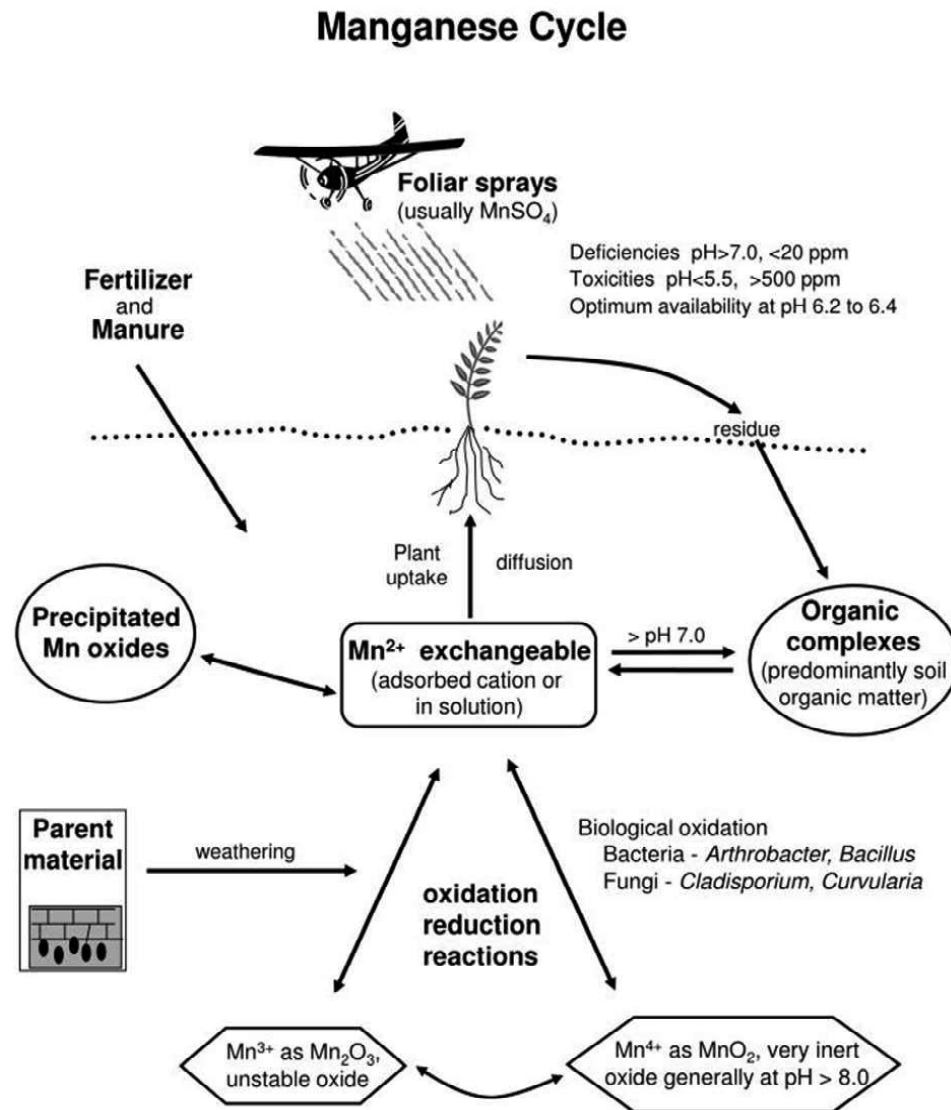


Fig. 2.19 Manganese Cycle

Figure 2.19 is depicting the manganese cycle.

Check Your Progress

1. Define biogeochemical cycle.
2. Which essential elements are added to the ecosystem by rock weathering?
3. How the carbon does returns back to the atmosphere?
4. What is nitrogen fixation?
5. How silica does enters into the aquatic environment?

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2.4 AQUATIC MICRO - ORGANISMS

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Aquatic microorganisms include members of the bacteria, protozoa, fungi and plant kingdom. These organisms differ radically, and share only their small size; most are not visible without a microscope, though colonies of some can be seen with the naked eye. Microorganisms are present in large quantities everywhere and can survive extreme physical and chemical conditions. Many microorganisms play foundational roles in aquatic ecosystems, capturing the sun's energy through photosynthesis and, through their role in decomposition, releasing nutrients stored in organic tissue. The microbiota present in aquatic bodies are as follows:

(a) Bacteria

- Kingdom Monera (Monos – Single) includes prokaryotes.
- Members of Kingdom Monera includes unicellular organisms (but one group is mycelial).
- Naked circular DNA is the genetic material in these organisms
- A nuclear envelope is absent.
- Both, ribosomes as well as simple chromatophores, are the only subcellular organelles in the cytoplasm.
- Sap vacuoles are absent.
- Instead, gas vacuole may be present.
- The predominant mode of nutrition is absorptive; however, few groups are photosynthetic (holophytic) as well as chemosynthetic in nature
- The organisms are non-motile or move by the beating of simple flagella or by gliding.
- Monerans augment the soil with nitrogen and hence serve as a significant part of the nitrogen cycle.
- Monerans are also very helpful in the production of some food items as well as antibiotics.
- Methanogens play an essential role in the treatment of sewage.
- Members of Kingdom Monera depends upon archaebacteria as the source of food.

Classification of Monera

Kingdom Monera is classified into three sub-kingdoms namely- Archaebacteria, Eubacteria, and Cyanobacteria.

Archaebacteria

- Archaebacteria are the most ancient bacteria found in the most extreme habitats such as salty area (halophiles), hot springs (thermoacidophiles) as well as marshy areas (methanogens).
- As they are ancient bacteria, the structure of the cell wall is different from that of the other bacteria which helps them survive in extreme conditions.

- The mode of nutrition is autotrophic in Archaeobacteria
- The nucleotide sequences of its t-RNA and r-RNA is unique.

Eubacteria

- Eubacteria are also referred to as “true bacteria”.
- The cell wall of Eubacteria is highly rigid and composed of peptidoglycans.
- Eubacteria moves with the help of flagella.
- Short appendages present on the cell surface of few Eubacteria are known as pili. These pili help the bacteria during sexual reproduction.
- Eubacteria are divided into two categories namely: gram-positive and gram-negative; dependent on the nature of the cell wall as well as the stain they take. The gram-positive bacteria retain the crystal violet colour and stains purple whereas the gram-negative bacteria lose crystal violet and stain red. Thus, the two types of bacteria are distinguished by gram staining.
- Examples of Eubacteria are: Rhizobium and Clostridium

Cyanobacteria

- Cyanobacteria are also referred to as blue-green algae.
- Cyanobacteria are photosynthetic in nature i.e. they are the producers
- Cyanobacteria contain chlorophyll pigments like carotenoids as well as phycobilins.
- A few Cyanobacteria are known for fixing atmospheric nitrogen.
- Nostoc, Anabaena, Spirulina are some of the best examples of cyanobacteria.

Characteristics of Bacteria

- Bacteria are microscopic organisms that can thrive well in diverse environments.
- Bacteria can be extremely useful for various commercial purpose as well as pathogenic too
- Naked circular DNA is the genetic material in these organisms
- A nuclear envelope is absent.
- Both, ribosomes as well as simple chromatophores, are the only subcellular organelles in the cytoplasm.
- The bacteria are surrounded by two protective coverings- the outer cell wall and the inner cell membrane. However, a few bacteria like Mycoplasma lack cell wall.
- A few bacteria are also covered by a capsule.
- Short whip-like appendages referred to as pili surround the surface of the bacteria which aids in carrying out sexual reproduction
- The long whip-like structures are known as flagella.
- Bacteria possess both autotrophic as well as heterotrophic mode of nutrition.

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- Autotrophic bacteria derive their nutrition from inorganic substances i.e. like green plants they are capable of synthesizing organic food/substance from inorganic material
- Heterotrophic bacteria depend entirely upon external organic materials as their source of food as they cannot synthesize their own food like autotrophic bacteria.

(b) Fungi

- Fungi are eukaryotic, non-vascular, non-motile and heterotrophic organisms.
- Fungi thrive well in moist and warm conditions.
- All the members of Kingdom Fungi contain a cell wall.
- Fungi are heterotrophic in nature i.e. they are dependent upon others for their nutrition.
- Fungi store their food in the form of starch.
- Fungi are widespread in nature.
- Fungi are used in the commercial production of beverages like wine and beer
- Some fungi are parasitic and can infect the host.
- Fungi are responsible for causing skin infections.
- Fungi are eukaryotic,
- The fungi have no embryonic stage. They develop from the spores.
- Fungi show the phenomenon of alternation of generation.
- Fungi lack chlorophyll.
- They cannot synthesize their own food via the process of photosynthesis and are dependent upon other for their nutritional requirements.
- Examples include mushrooms, moulds, yeast.

Fungi can be both beneficial as well as harmful.

- o **Recycling** – Fungi plays an essential role in recycling dead and decaying organic matter.
- o **Food** – Fungi like mushrooms are edible and are used for human consumption.
- o **Medicines** –Fungi are used for developing antibiotics to control diseases in humans as well as animals. One of the common antibiotic Penicillium is derived from a common fungus.
- o **Biocontrol Agents** – Fungi help in controlling pests like insects and other small worms.
- o **Food spoilage** – Fungi are responsible for spoiling the food like bread.
- o **Skin infections**- Fungi are responsible for severe skin infections which might be fatal sometimes
- o **Beer and Wine Production**- Fungi are used in the commercial production of beer and wine.

Structure of Fungi

- Majority of the members of kingdom Fungi have a filamentous structure except the yeast cells.
- Fungi can either be single-celled or multicellular organism.
- Fungi are composed of long thread-like structures known as hyphae.
- These hyphae combine together to form a mesh-like structure known as mycelium.
- The cell wall of Fungi is composed of chitin and polysaccharides.
- The nucleus is dense, clear, with chromatin threads.
- The nucleus is surrounded by a nuclear membrane.

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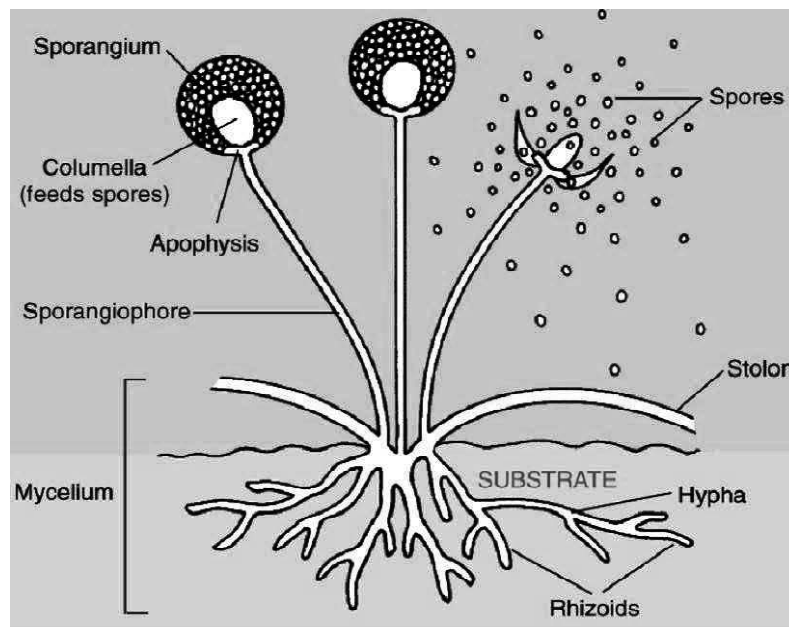


Fig. 2.20 Structure of Fungi

Figure 2.20 depicting the structure of fungi.

(c) Protozoa

- Protozoans are unicellular or acellular organisms. The body comprises of mass of protoplasm, hence they are called as acellular animals. A single cell performs all the vital metabolic functions of the body.
- Protozoa are eukaryotic microorganisms.
- They are notable for their ability to move independently, a characteristic found in the majority of species.
- Protozoans are located in most habitats; free-living species inhabit freshwater and marine environments, whereas terrestrial species inhabit decaying organic matter. Some species are parasitic in nature.
- Protozoans vary substantially in size and shape. Smaller species are of the size of fungal cells; larger species may be visible to the unaided eye.

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- Protozoan cells lack cell walls and therefore can take an infinite variety of shapes. Body of the organisms is either naked or covered with a structure known as pellicle
- Most protozoan species are aerobic, but some anaerobic species have also been found in the human intestine and animal rumen. No specialized respiratory structures. It occurs through the general body surface
- Protozoans are **heterotrophic** microorganisms, and most species obtain large food particles by **phagocytosis**. The food particles are ingested into a food vacuole and lysosomal enzymes then digest the nutrients in the particle, and the products of digestion are distributed throughout the cell. Some species have specialized structures known as **cytostomes**, through which particles pass inside.
- Many protozoans alternate between a free-living vegetative form known as **trophozoite** and a resting form called a **cyst**. Many protozoan parasites are taken into the body in their cyst form. The protozoan cyst is somewhat analogous to the bacterial spore, since it resists harsh environmental conditions.
- Most protozoal species move independently by one of three types of locomotor organelles: flagella, cilia, and pseudopodia. **Flagella** and **cilia** are structurally similar, having a “9-plus-2” system of microtubules, the same type of structure found in the tail of animal sperm cells and certain cells of unicellular algae. How a protozoan moves is an important consideration in assigning it to a group.
- Protozoans play an important role as **zooplankton**, the free-floating aquatic organisms of the oceans, they are found at the bases of many food chains, and participate in many food webs.
- Most protozoans have a single nucleus, but some have both a macronucleus and a micronuclei. Contractile vacuoles may be present in protozoans to remove excess water, and food vacuoles are often observed.
- A **Contractile Vacuole (CV)** previously known as **pulsatile** or **pulsating vacuole** is a sub-cellular structure or organelle which is primarily involved in the process of osmoregulation. It is observed chiefly in protists and unicellular algae. A contractile vacuole is formed when a large number of tiny droplets fuse in the area where the contractile vacuole is to be formed as in amoeba or around the mitochondria contractile vacuole as in Euglena. The energy required for the functioning of contractile vacuole is provided by the mitochondria surrounding the vacuole. Contractile vacuoles osmoregulate by removing excess water from the body. It's also believed to be excretory in function. In freshwater forms, body fluid is hypertonic to the surrounding medium. Being surrounded by water dominant media, water continuously enters the body of freshwater protozoans increasing the internal hydrostatic pressure. This inhibits normal functioning and after a threshold the body may burst. Thus, by removing excess water, contractile vacuole maintains the internal hydrostatic pressure. However, in marine and parasitic

protozoans, the media surrounding them is isotonic such that no water enters the body. Thus, these forms don't require a contractile vacuole.

- Although most protozoans reproduce by asexual methods, sexual reproduction has been observed in several species.

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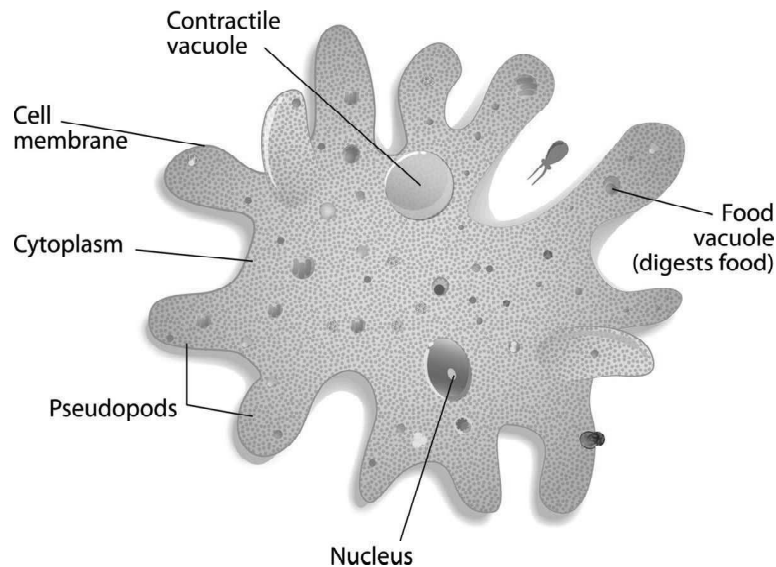


Fig. 2.21 Structure of Simplest Protozoan

Figure 2.21 is depicting the structure of simplest protozoan, i.e., Amoeba.

(d) Algae and Phytoplankton

Phytoplankton are small microscopic, unicellular and photosynthetic organisms which freely float in water bodies. Phytoplankton composed of both eukaryotic as well as prokaryotic species which colonizes upper euphotic part of the water column ranging from freshwater to ocean conditions. They also exhibit amazing adaptation to remain in floating condition. The range of the Phytoplankton cells can vary in size from about 1 μm to 1 mm. Like terrestrial plants, these tiny primary producers need sunlight, nutrients as well as carbon dioxide for their growth & multiplication. Phytoplankton cells contain chlorophyll which helps them to synthesize food in the presence of sunlight. Therefore, they help in maintaining not only the carbon dioxide level but also help to alleviate global warming to some extent. Physical processes like wind and current play a substantial part in their distribution particularly in estuarine and marine conditions. Phytoplankton act as a primary link in the energy pathway to higher trophic levels via several food chains. Phytoplankton supports nearly half of global primary production which directly or indirectly supports almost all marine life. Phytoplankton are a chief food source for a diverse range of organisms like zooplankton, larvae, and juveniles of fishes as well as some invertebrates. The annual cycle of the phytoplankton remains predictable throughout the year, however, occasionally some species may develop exponentially leading to the formation of algal blooms. Eutrophication refers to the process in which a water body becomes overly enriched with nutrients, leading to excessive growth of algae and plankton. The excessive growth (or bloom) of algae and plankton in a water body are indicators of this process. Eutrophication is considered to be a serious environmental matter as it often leads to the deterioration of water

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quality and the depletion of dissolved oxygen (DO) in aquatic bodies. Eutrophic waters can eventually become “dead zones” that are no longer capable of supporting aquatic life (both plants and animals). Over a period of time, anthropogenic activities like use of excessive fertilizers in agricultural field, disposal of nutrient rich waste into water bodies etc. has increased the nutrient content of freshwater bodies ultimately leading to the deterioration of water quality and making it unfit for human consumption.

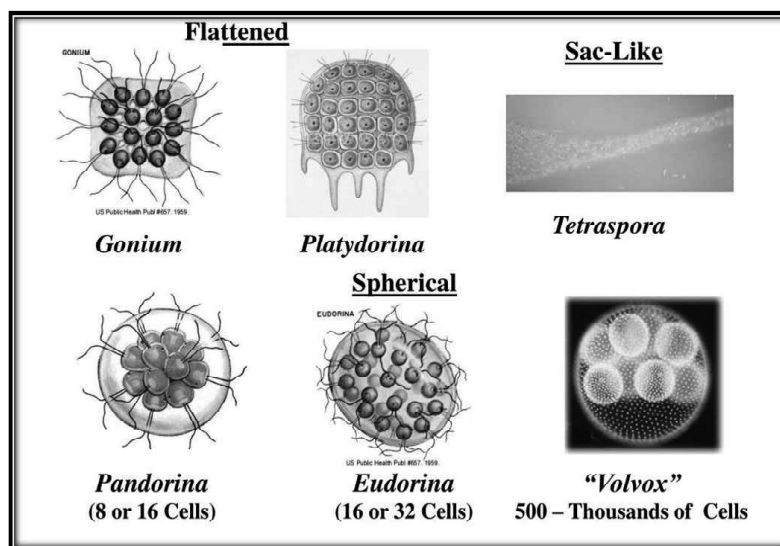


Fig. 2.22 Phytoplankton

Figure 2.22 is depicting the phytoplankton.

Phytoplanktons can be:

Microplankton – Size range varies from 200-20 μm ;

Nanoplankton- Size range varies from 20-2 μm ;

Picoplankton- Size range varies from 2-0.2 μm .

(e) Zooplankton

The name zooplankton is derived from the Greek: Zoon, animal; planktos, wandering. Zooplankters are microscopic, unicellular or multicellular forms with size ranging from a few microns to a millimeter or even more. Apart from size variations, zooplankters exhibit differences in morphological features as well as taxonomic positions consisting of representatives from almost every taxon of the animal kingdom. They are diverse, gentle and beautiful groupings of animals that drift in the aquatic bodies and add substantially to the faunal biodiversity of aquatic bodies. They play a significant role in the marine food web. They help in transferring organic energy obtained from unicellular algae to higher trophic levels like pelagic stocks. They act as food source for larval as well as juvenile fish, hence their reproductive cycles, growth as well as survival rates are essential factors influencing fishery.

Fresh-Water Microbiota

A variety of microorganisms live in different zones of lentic freshwater. The region of a water body near the shoreline is known as littoral zone. This zone consisting

of shallow and warm waters is present around the margins of the aquatic body. Plenty of light is available in this zone and supports the growth of photosynthetic algae as well as bacteria that use light as energy thrive in this zone. Further away from the shore is the limnetic zone. Limnetic zone is the zone of open waters which are deeper as well. Availability of plenty of light in this zone promotes active photosynthesis as well as growth of free-floating autotrophs like planktons. Phytoplankton and periphyton are the chief primary producers in lentic water system. Phytoplankton are seen drifting in the water column of the pelagic zone. However, several phytoplankton have a higher density and sink to the benthic region. Periphytons remains attached to the substrate covering almost the entire benthic surfaces. Phytoplankton as well as periphyton serves as important food source and chief oxygen producers in the lentic system. In a lake system, the major phytoplankton of the littoral zone are holophytic blue green algae (*Microcystis*, *Oscillatoria*) flagellates diatoms and green algae (*Cosmarium*, *Staurastrum*). In a Lake system, zooplankton like *Dysticus*, *Stentor*, *Vorticella*, *Laccotrohes* have been observed Further, Zooplankton in the littoral region are water flea such as *Daphnia*, *Moina* etc. The nektons found are *Euglena Coroxa*, *Dysticus*, *Paramecium*, *Gyrinus*, *Chaoborus*, *Ranatra*. The community of the limnetic zone (lakes) is mostly composed only of plankton, nekton, and sometimes neuston (organisms resting or swimming on the surface). Phytoplankton producers consist of diatoms, green algae, blue- green algae, and algae- like green flagellates, chiefly the dinoflagellates. Many microscopic creatures like *Volvox*, *Euglena*, *Phacus* have been observed in limnetic zone of a lake system.

Similarly, in a pond ecosystem, Phytoplankton like *Oscillatoria*, *Anabaena*, *Eudorina*, *Volvox* and Diatoms are present abundantly. Further, filamentous algae such as *Ulothrix*, *Spirogyra*, *Cladophora* and *Oedogonium* can also be seen. Primary consumers of the pond ecosystem are zooplanktons like *Paramoecium* and *Daphnia*. Photosynthetic microbes also live here in both littoral as well limnetic zones of lentic water system. Profundal zone is present beneath the limnetic zone and receives very less light. Hence, this zone is known as aphotic zone in contrast to euphoric zone (limnetic and littoral regions) which are well illuminated. As the water deepens, temperatures become colder and the oxygen concentration as well as light in the water decrease. Now, microbes that require oxygen do not thrive. Instead, purple and green sulfur bacteria, which can grow without oxygen, dominate. Finally, benthic zone is present beneath the profundal zone. Benthic zone is present near the bottom of the aquatic body. Profundal as well as benthic zones are characterised by presence of heterotrophs which survive on dead and decaying organic material coming from limnetic zone present above. Few microbes survive in this zone. Bacteria that can survive in the absence of oxygen and sunlight, such as methane producing bacteria, thrive. Profundal zone is mostly inhabited by presence of fungi as well as some bacteria most of them are anaerobic in nature. These animals can survive easily in low light as well as low levels of oxygen. Autotrophic organisms fail to thrive in this area due to lack of sunlight. Their major source of energy is dead and decaying organic matter coming from the limnetic zone. All the organisms found in the profundal region are parasitic, carnivores or detritivores in nature. For instance: - Decomposers are present in mud water and bottom of the ponds and helps in the

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recycling of nutrients by carrying out the process of decomposition. For instance: Bacteria and Fungi are decomposers. Bacteria are present in all regions of lentic waters (lake, pond etc.). Free-living bacterial forms are associated with decomposing organic material, or are present on the surfaces of rocks as well as plants, suspended in the water column, and in the sediments of the benthic & profundal zones. A few other bacterial forms are associated with guts of lentic animals as parasites or in commensal relationships. Bacteria play an essential function in system metabolism via nutrient recycling. Majority of bacteria in lentic water system depends upon dead and decaying organic matter to obtain their energy. These bacteria are then eaten up by protozoa which in turn are consumed by zooplankton and then further up the trophic levels. Zooplankters are microscopic, unicellular or multicellular forms with size ranging from a few microns to a millimeter or even more. Apart from size variations, zooplankters exhibit differences in morphological features as well as taxonomic positions consisting of representatives from almost every taxon of the animal kingdom. Zooplankton exhibits diverse food habits. For instance: - Microscopic zooplankton Protozoa feed upon minute algae and bacteria. Utilisation of dissolved substances has been demonstrated in certain protozoa.

Lotic aquatic systems are those systems which contain flowing waters. For instance: Springs, streams and rivers are familiar examples of lotic systems. The primary function of these lotic bodies of water is to carry the surplus rain water back to the sea. Lotic water system exhibits longitudinal zonation or profile instead of horizontal as seen in lakes and ponds discussed in the previous sections. Bacteria are present in all regions of lotic waters. Free-living bacterial forms are associated with decomposing organic material, or are present on the surfaces of rocks as well as plants, suspended in the water column, and in the sediments of the benthic and profundal zones. A few other bacterial forms are associated with guts of lentic animals as parasites or in commensal relationships. Bacteria play an essential function in system metabolism via nutrient recycling. Majority of bacteria in lotic water system depends upon dead and decaying organic matter to obtain their energy. These bacteria are then eaten up by protozoa which in turn are consumed by zooplankton and then further up the trophic levels. Algae, comprised of phytoplankton and periphyton, are the major primary producers in most streams and rivers (Giller and Malmqvist 1998). Phytoplankton float freely in the water column however they are unable to maintain high populations in fast flowing streams. Phytoplankton population can rise steadily in slow moving rivers and backwaters (Allan 1995). Periphyton are typically filamentous in structure and remain attached to the objects to avoid being washed away by fast river/stream current. In slow moving rivers/streams, periphyton may form a gelatinous, unanchored floating mat (Cushing and Allan 2001). Phytoplankton as well as periphyton are essential to lotic systems and serve as food source, sources of energy, microhabitats for several animals, etc.

Biotic communities of the rhithron zone comprises of plankton, periphyton, “aufwuchs”, nekton as well as benthos. They are described as follows:

- (a) **Phytoplankton:** Phytoplankton are the major primary producers in the ‘Rhithron’ zone. Phytoplankters are small microscopic, unicellular and photosynthetic organisms which freely float in water bodies. Phytoplanktons

composed of both eukaryotic as well as prokaryotic species which colonizes upper euphotic part of the water column. In rhithron zone of comparatively undisturbed stream, the major primary producers are the filamentous green algae like *Ulothrix* and *Cladophora* and diatoms like *Nitzschia* and *Gomphonema*. *Ulothrix* belongs to the class Chlorophyceae. They are usually green due to the presence of pigments like chlorophyll a, chlorophyll b and beta-carotene. Pigments are present in plastids or chromatophores. Starch is the main photosynthetic food product. In chromatophores pyrenoids are present. Both flagella are equal in length i.e. they are isokontate. Cell wall is made up of cellulose. Diatoms are members of the class Bacillariophyceae (division Chromophyta), with approximately 16,000 species found in sediments or attached to solid substances in aquatic bodies. The markings of cell wall, structure and position of raphae and nodules are the distinguishing features for identification of species. The diatom cell is well known as frustules and typical feature is possession of silica cell wall. This structure is extremely ornamental, which is species specific and frequently used as means of identification. Diatoms are among the most significant and productive microscopic sea organisms and serve directly or indirectly as food for many animals.

- (b) **“Aufwuchs”**: Those organisms that attach firmly to a substrate (submerged rocks, plants, debris) but do not penetrate it (in contrast to plants rooted in the bottom or certain parasites) are collectively called “known as “Aufwuchs””.
- (c) **Zooplankton**: The zooplankton of the ‘Rhithron zone’ majorly comprises of ciliate protozoans (For instance:- *Vorticella*) Rotifers (For instance:- *Asplancha*, *Brachionus*, *Keratella*, *Philodina*), and crustaceans (For instance:- *Daphnia*, *Bosmina*, *Cyclops*, *Diaptomus*, *Gammarus pulex*).

The biotic communities of Potamon zone are as follows:

- (a) **Plankton**: The presence of plankton is associated with the flow conditions. During floods, rare planktonic organisms may be present however during dry seasons algal blooms may develop within the lentic waters of the plain as well as in the main channel. In short rivers, these are usually confined to backwaters whereas in extended rivers the time taken for specific masses of water to travel downstream is enough to let the development of plankton.
- (b) **Algae and Diatoms**: Algae and Diatoms are the most common phytoplankton present in potamon zone of streams. The type of algal species growing in potamon zone depends upon its tolerance to pollution. For instance, algal species like *Euglena viridis*, *Scenedesmus quadricauda*, *Microcystis*, *Oscillatoria limosa*, *Ankistrodesmus falcatus*, *Phacus caudatus*, *Navicula viridula*, *Stigeoclonium tenue*, *Chlorella vulgaris*, *Synedra ulna* and *P. viridis* are regularly limited to the sites of urban & industrial discharge. However, algal species like *Anabaena circularis*, *Gomphonema parvulum*, *Phormidium uncinatum*, *Pediastrum duplex*, *Cymbella turgida*, *Oscillatoria princeps*, *Scenedesmus dimorphus*, *Closterium acerosum*, *Synedra acus*, *Navicula cuspidate* are pre-dominantly present in clean or moderately polluted zones. Further, algal species like *Oscillatoria subbrevis*, *Melosira ambiguans*, *Pediastrum simplex*,

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Phormidium calcicole, *Merismopedia glauca*, *Gomphonema* species are present in highly clean water.

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- (c) **Zooplankton:** Zooplankton members of the potamon zone comprises of members like copepoda, protozoa, rotifera Cladocera. Further, species like *B. quadridentate*, *B. forficula*, *B. calyciflorus*, *Filinia longiseta*, *K. cochlearis*, *Brachionus angularis*, *Keratella tropica* are present in both clean as well as moderately polluted waters. However, species like *Rotatoria rotatoria* *Platijas polyacanthus* *Brachionus rubens* are representative of polluted parts of the potamon zone.

Sea-Water Microbiota

Seawater, or salt water, is water from a sea or ocean. On an average, seawater in the world's oceans has a salinity of about 3.5% (35 g/l, 35 ppt, 600 mM), i.e., roughly one liter of seawater has approximately 35 grams of dissolved salts (predominantly sodium (Na⁺) and chloride (Cl⁻) ions). Average density at the surface is approximately 1.025 kg/l. Seawater is denser than both fresh water and pure water (density 1.0 kg/l at 4 °C (39 °F) as the dissolved salts increase the mass by a larger proportion than the volume. The freezing point of seawater decreases as salt concentration increases. At typical salinity, it freezes at about -2 °C (28 °F). The pH of the seawater is typically limited to a range between 7.5 and 8.4. However, as such, there is no universally accepted reference pH-scale for seawater and the difference between measurements based on different reference scales may be up to 0.14 units. Saltwater presents a very different environment to microorganisms. The higher salt concentration, higher pH, and lower nutrients, relative to freshwater, are lethal to several microorganisms. However, halophilic bacteria (salt loving bacteria) abound near the surface, and some bacteria that also live-in freshwater are plentiful (For instance- *Pseudomonas* and *Vibrio*). Also, few research have documented that the ancient form of microbial life known as archaeobacteria is one of the predominant forms of life in the ocean. Archaeobacteria are the most ancient bacteria found in the most extreme habitats such as salty area (halophiles), hot springs (thermoacidophiles) as well as marshy areas (methanogens). The role of archaeobacteria in the ocean food chain is not yet known, but must be of vital importance. Another microorganism found abundantly in saltwater are a type of algae known as dinoflagellates. The rapid growth and multiplication of dinoflagellates can turn the water red. This "red tide" depletes the water of nutrients and oxygen, which can cause many fish to die. As well, humans can become ill by eating contaminated fish.

Check Your Progress

6. How the microorganisms play foundational roles in aquatic ecosystems?
7. Which are the most ancient bacteria found in the most extreme habitats such as salty area?
8. Give some examples of cyanobacteria.
9. What do you understand by the term phytoplankton?
10. Which zone of a water body is present near the shoreline.

2.5 ROLE OF MICROORGANISM IN WATER PURITY TEST

Water can support the growth several organisms. In some cases, it is advantageous as certain yeasts can provide good quality of bread and beer. However, some microorganisms are unhealthy and even life threatening. Some of these incidents have resulted in serious, widespread sickness, even death. Presence of microbial pathogens or contaminants in drinking water are being blamed for several gastrointestinal illnesses that have occurred worldwide. Now waterborne sickness from microbial contaminants, like *Cryptosporidium*, *Giardia*, *Legionella* and Norwalk virus-has become a seemingly modern concern even for people living in the advanced nations like United States. The Centers for Disease Control and Prevention estimates 900 to 1,000 people die each year from microbial illnesses from U.S. drinking water. Other estimates run as high as 1,200 deaths. Although difficult to pin down, such figures indicate the existence of a serious problem.



Fig 2.23 Microorganism in Water

Microorganisms are present everywhere in our environment, i.e., in soil, air, food as well as in water. Microorganisms which are also referred to as microbes are living organisms, generally observable only via a microscope. Our exposure to them causes harmless microbial flora to establish in our bodies, even though some microbes are pathogens and can cause serious water borne diseases. These diseases are considered waterborne only if the pathogens are transmitted by water, to infect humans or animals that ingest the contaminated water. Diseases transmitted by water are primarily those found in the intestinal discharges of humans or animals. The presence of microbial contaminants in drinking water has plagued humans throughout history. Infectious diseases spread from infected person to healthy ones via several routes like air, food, water, etc. Water borne diseases are referred to as those diseases that spreads via drinking or using contaminated water. Water generally gets contaminated under poor hygienic or in-sanitary conditions. Contamination can occur either at the source of water supply, while passing through water pipes, which are broken, or at homes too when water is not stored properly. The water can be contaminated with either human or animal faeces and the diseases transmitted by drinking such water are known as faeco-orally transmitted diseases.

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Water borne diseases can also be transmitted through eating food prepared using contaminated water or using utensils which have been washed with the contaminated water.

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The two major source of infection are:

(1) Drinking Water: Drinking water can be contaminated via:

- Contamination at its main source
- Contaminated during supply or storage of the water.
- Ice made from contaminated water.

(2) Food: Food can be contaminated via:

- Contaminated during or after preparation of the food.
- Using utensils washed with contaminated water for food preparation
- Fruits and vegetables, washed with contaminated water and eaten raw.
- Using Fruits and vegetables, grown at or near ground level and irrigated with water contaminated with human waste, and eaten raw.

Water-borne diseases are widespread in areas having:

- Insufficient supply of water
- Poor quality of water
- Problematic or badly laid sewage pipelines
- Poor sanitary conditions
- In remote villages where water from step wells or uncovered wells or ponds are used for multipurpose like bathing, washing clothes as well as for drinking.
- Defecation in the open particularly near major source of drinking water
- Poor system for the proper disposal of human waste
- Water-borne diseases can also occur if contaminated water is used for Kitchen purpose like washing utensils or for washing fruits and vegetables especially when eaten raw.
- Water borne diseases can also be transmitted by using ice prepared with low quality water from unreliable sources.

Though, water borne diseases may occur in individuals throughout the year yet a seasonal increase is generally noted in specific season like summer, monsoon or post-monsoon period. Over a period of time, it has been observed that the risk of large outbreaks of water-borne diseases surges following heavy rains. Waterborne microbial pathogens are responsible for causing a whole range of diarrheal diseases. The hazards of fecal contamination as well as the principles of basic sanitation were recognized early. The occurrence of such outbreaks alerted people to the hazards of drinking contaminated water and prompted investigations into ways to prevent the occurrence of waterborne illnesses. Public health officials eventually achieved success in controlling the more common forms of waterborne diseases, at least in the United States and other developed countries. Progress was due to the adoption of public health measures as well as the implementation

of important water treatment techniques, such as filtration, disinfection and sewage treatment. Some believed the battle, if not won, was at least under control.

All these microbial contaminants are responsible for causing water borne diseases like: Typhoid Fever whose Infective agent is *Salmonella typhi* (Bacteria). Cholera is another highly infectious water borne disease whose infective agent is *Vibrio cholerae* (Bacteria). Serogroup O1 and O139- are responsible for causing cholera.

Waterborne microbial contaminants, however, have attracted renewed attention, both within the scientific community as well as among the general public. There was a time, when it was considered to be under control. However, in the present era, they are now referred to as the “emerging drinking water contaminants.” What in fact is emerging is an expanded awareness of the presence of previously undetected microbial contaminants in drinking water and their effects on human health. Moreover, new microbial pathogens are being discovered and research is underway to develop improved methods for detecting and treating microbes in drinking water. For instance; Microsporidia is an example of an emerging pathogen that is attracting attention. Potentially waterborne, this pathogen is recognized as causing disease among AIDS patients, although healthy persons also may be susceptible to microsporidia. Due to its extremely small size, microsporidia may thrive the process of filtration, and the research studies thus far specify that the pathogen will be justly resistant to many drinking water disinfectants. With more research as well as the development of improved detection methods, researchers will be able to better determine the occurrence of microsporidia, both in humans and the environment. A few researchers believe this microorganism might eventually need to be monitored and controlled in drinking water supplies. *H. pylori* is another emerging pathogen. *H. pylori* is certainly common in people who have been exposed to poor hygienic conditions since childhood. *H. pylori* also has been observed, even though irregularly, among the socioeconomic advantaged. *H. pylori* causes severe inflammation of the stomach and is seen as one of the important factors responsible for the development of duodenal ulcers. It has also been suggested that it might be playing some role in causing gastric cancer. The occurrence of *H. pylori* ranges from less than one percent of the population of industrialized nations to three to eight percent in developing countries. Researchers continue to study this pathogen. These as well as other microbial contaminants are increasingly posing a great threat to the human health and hence attracting the concern of public health authorities as well as an interdisciplinary array of experts in the fields like microbiology, engineering, epidemiology and risk assessment. As we have seen in the previous section, several water borne diseases like typhoid, cholera etc. result from drinking fecal contaminated water. In order to explain the presence of microbial contaminants in drinking water it is required to describe a circuitous route, from a human or animal source back to a human or animal via drinking water. Microbial contaminants follow a fecal-oral route. Bacteria, viruses, as well as protozoa are the microorganism groups containing pathogens of primary concern in the study of waterborne disease.

Human sources account for viruses, on the contrary, both animal as well as human waste contribute protozoa to water. For instance, cattle are regarded to be

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the source of much *Cryptosporidium*, and *Giardia* is usually traced to beavers. Both *Cryptosporidium* as well as *Giardia* are protozoa.

Every single day the average human excretes approximately 38 grams of urea, primarily urine, and 20 grams of solids in feces. The excreta comprises of billions of microbes. These microbes cannot only thrive, but are also capable of multiplying in water and are made up of a broad range of organisms, including pathogenic microbes, which even healthy people excrete. Others individuals who are suffering from a water borne disease or who are carriers of a disease-producing microorganism are a more obvious source of waterborne infections. Approximations specify that approximately five percent of those who have contacted an enteric or intestinal disease remain life-long carriers of that particular disease, even if they recovered fully from the disease. That these intestinal microbial contaminants can infect a drinking water source may at first seem puzzling, especially to citizens of a country that prides itself on its public health standards. Yet through natural flow or accident, various types of water can interconnect and flow together. For instance, stormwater runoff from residential, rural and urban areas can carry waste material from domestic pets and wildlife, to collect in surface waters and even enter groundwater. Through accident or equipment failure, sewage, a rich source of microbial contamination, might come into contact with drinking water.

Further, defective on-site wastewater disposal or septic systems in rural as well as other residential areas can contribute huge number of coliforms and other bacteria which are responsible for causing both waters borne diseases to both surface water and groundwater. These microbial contaminants occur widely and are not limited to areas that are inhabited by humans.

Water purity test

An important aspect of water microbiology, particularly for drinking water, is the testing of the water to ensure that it is safe to drink. Thus, it is essential to study microbial contaminants as they are responsible for causing several water borne diseases of public health importance. Water borne diseases are regarded to be of public health importance due to the following reasons:

- Water borne diseases have the potential for causing large outbreaks.
- High disease burden owing to large number of hospital cases and deaths, especially in young individuals.
- In many Indian states as well as districts, water borne diseases is chief causes of pediatric indoor admissions as well as outpatient visits to the hospitals.
- No specific treatment is available for some viral diseases such as hepatitis
- Increasing anti-microbial drug resistance is being recorded for some water borne bacterial diseases.
- Water borne diseases have the potential for causing panic in the community as well as negative media coverage.

Microbiological water testing looks for indicator organisms as a sign of faecal contamination rather than testing for specific pathogens. Correct sampling procedures as instructed by international standard protocols must be adhered to

routine microbiological testing of drinking water supplies, recreational waters, and environmental waters is essential for the protection of public health. Water quality testing can be done in several ways. A few ways are discussed below:-

(1) Testing of indicator organisms

One of the most effective way to check water supplies for faecal contamination is microbiological analysis, and a range of test methods designed for that purpose has been developed for the water industry. Instead of carrying out individual tests for the potential pathogens like viruses, or parasites that might be in the water, microbiologists test for indicator organisms that are always present when enteric pathogens and viruses are. Coliform is a term which is used to specify a group of gram-negative bacteria that can ferment lactose with a production of gas within 48 hours at a temperature from 35°C or 44/44.5°C. These features allow for easy isolation, detection, as well as listing in the lab and are the gold standard for the microbial testing of water. They are always present when enteric pathogens or viruses are detected in water testing. However, a high 'total coliform' count doesn't always mean faecal contamination and needs an alternate method to identify the faecal coliforms from coliforms found in the environment. *Escherichia*, *Enterobacter*, *Klebsiella* are the faecal coliforms, and *Citrobacter* and *Serratia* are found in plants and soil. The faecal coliforms, are also referred to as thermotolerant coliforms which can thrive at a temperatures of 44°C and 44.5°C. This temperature allows simple differentiation between the two types of coliforms. The researchers can observe for counts of faecal coliforms like *E. coli*, whose only habitat is the intestine, and whose life outside the gut or intestine is very short-lived; is seen as the ideal indicator organism. The presence of *E. coli* in a sample of drinking water indicates that the water is contaminated with microbial impurities and hence is unsafe for human consumption.

The presence of faecal streptococci/*Enterococci* is evidence of faecal contamination. Faecal streptococci tend to continue for longer duration in the environment as compared to thermotolerant or total coliforms that are extremely resilient to drying. Hence, it is likely to isolate faecal streptococci from water that consists of few or no thermotolerant coliforms as, for instance, when the source of contamination is distant in either time or space from the sampling point. Faecal streptococci grow in or on a medium containing sodium azide, at a temperature of 37-44 °C. They are generally noticed by the drop in the amount of a dye (generally a tetrazolium-containing compound) or the hydrolysis of aesculin. Traditional techniques may give "false positives," and other supplementary confirmatory tests may be required.

(2) Rapid Methods

Traditional methods of microbial detection tend to be labor-intensive and take more than a day to yield results. Rapid methods for microbial detection can be sensitive, precise, and quick. In general, rapid methods can be grouped into three distinctive categories in accordance with their application. These categories include qualitative, quantitative, and identification methods. Qualitative rapid methods provide a presence or absence result that indicates microbial contamination in a sample. Quantitative methods provide a numerical result that indicates the total

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number of microbes present in the sample. Identification methods provide us with a species or genus name for the microbial contaminant in a sample. Various rapid methods are available and in use in the industrial microbiological market today. They tend to be based on various technology platforms. The more common technologies include nucleic-acid-based detection, which uses DNA or RNA targets; antibody-based detection; biochemical; enzymatic detection such as adenosine triphosphate (ATP) methods; impedance methods; and flow-cytometry-based methods. Sanger sequencing (SGS) is the first-generation sequencing method which was developed by Fredric Sanger in the year 1977.

(3) Plating Techniques (Culture techniques)

A lot of selective media have been developed for the detection of indicator organisms in water by MF methods. For instance:- Recommended media for coliforms as well as *E. coli* include membrane lauryl sulphate broth or agar, MI agar and broth, and membrane lactose glucuronide agar. Membrane Enterococcus Agar (mEA) and membrane-Enterococcus Indoxyl- β -D-Glucoside Agar (mEI) can be used for detection and enumeration of enterococci, while Tryptose sulphite cycloserine agar without egg yolk can be used to culture *Clostridium perfringens* on membrane filters. *Pseudomonas aeruginosa* can also be detected by an MF method using Pseudomonas agar. Further culturing, or even biochemical testing can then be used to confirm the identity of suspect colonies growing on filters placed on selective media. Increasingly, chromogenic as well as fluorogenic media are frequently used in water microbiology. Based on the detection of specific enzymes in the target bacterial species by substrates containing chromogenic or fluorogenic groups, producing highly diagnostic-coloured colonies, these media can be less harsh than other selective media, resulting in fewer false-negative results, and reduce the time needed to confirm results. Techniques using pour and spread plate count methods are not highly sensitive techniques for the detection of indicator organisms as well as pathogens in water, even though, they are still used routinely for counting heterotrophic bacteria. Methods capable of testing a larger volume of water (typically 100 ml) are required. For several years, the primary method of choice was the multiple tube 'Most Probable Number' (MPN) technique, in this technique, the measured volumes of the water sample are added to a series of tubes containing differential media and incubated. Growth of the microorganism is indicated by a colour change in the medium, and the result is estimated from the distribution of positive tubes. Even though, the method is simple as well as inexpensive in terms of equipment and materials used, it is a laborious method and need huge amounts of incubator space. It is also an indirect method and does not allow further examination of individual colonies. MPN tests for routine water microbiology have now been largely replaced by Membrane Filtration (MF) methods, although they may still be useful for occasional tests conducted in small laboratories or the field, and commercial test kits based on MPN methods are available for coliforms and enterococci.

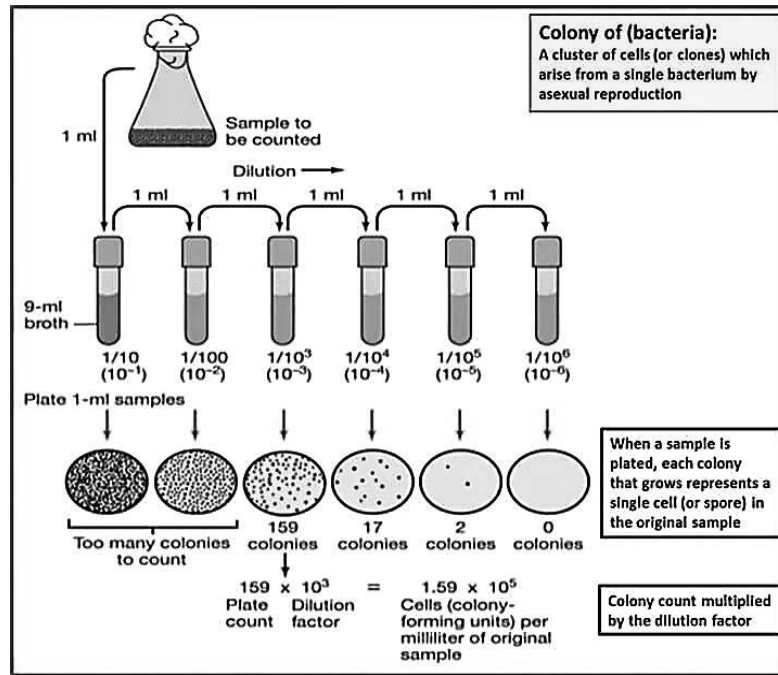


Fig. 2.24 Plating Technique

Figure 2.24 is depicting the plating technique.

(4) Sequencing methods

Prokaryotic ribosomes consists of two subunits namely large subunit having size 70s and small subunit having size 30s. 16srRNA is associated with smaller subunit of ribosome, whereas 23s rRNA and 5srRNA is associated with larger subunit. 16srRNA is nearly 1.6kb in length. The genes which codes for 16srRNA are referred to as 16srDNA.

Only 1% of bacterial population is culturable and can be isolated by plating on different media used in lab. The rest of the 99% is unculturable and cannot be isolated by traditional methods (Amann *et al.*, 1995). Hence there arises a need for a suitable marker for studying the rest of the bacterial population. 16srRNA due to its ubiquitous presence, significant evolutionary properties, highly conserved sequence with species specific signature sequence served as efficient marker in identification and taxonomic nomenclature of the newly identified bacterial species (Patel, 2001). The sequencing of the 16srRNA is carried out by using universal primers. The most common primers used are 27F and 1492R. A minimum of 500 bases is needed for taxonomically assigning the bacterial species. However 1300 to 1500 base sequence is required for categorizing the isolated bacteria as new species. The nearest neighbour indicating a similarity score of <97% represent new species (Petti *et al.*, 2007). However the quick and easy to use technique has some major shortcomings like reliability of the sequence generated. So DNA-DNA relatedness is still the most reliable method for definitive assignment of species. DNA-DNA hybridization values of less than 70% represent new species. Sanger sequencing involves the selective incorporation of chain terminating di-deoxynucleotides by DNA polymerase during *in vitro* DNA replication. The amplicons thus produced are separated from each other by the process of capillary

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electrophoresis. Usually, Sanger sequencing acts as a quick and cost-effective technique for low number of samples say less than 100. Further, it is better for the sequencing of single genes. Furthermore, as mention above too Sanger sequencing is an analogical method, i.e., it generates a single sequence by combining signals from all DNA fragments in the sample. Sanger sequencing does not allow the isolation of individual signals. Consequently, the resultant signal is a mixed-signal, which does not allow the identification of variants, which occur below 25% frequency in a sample.

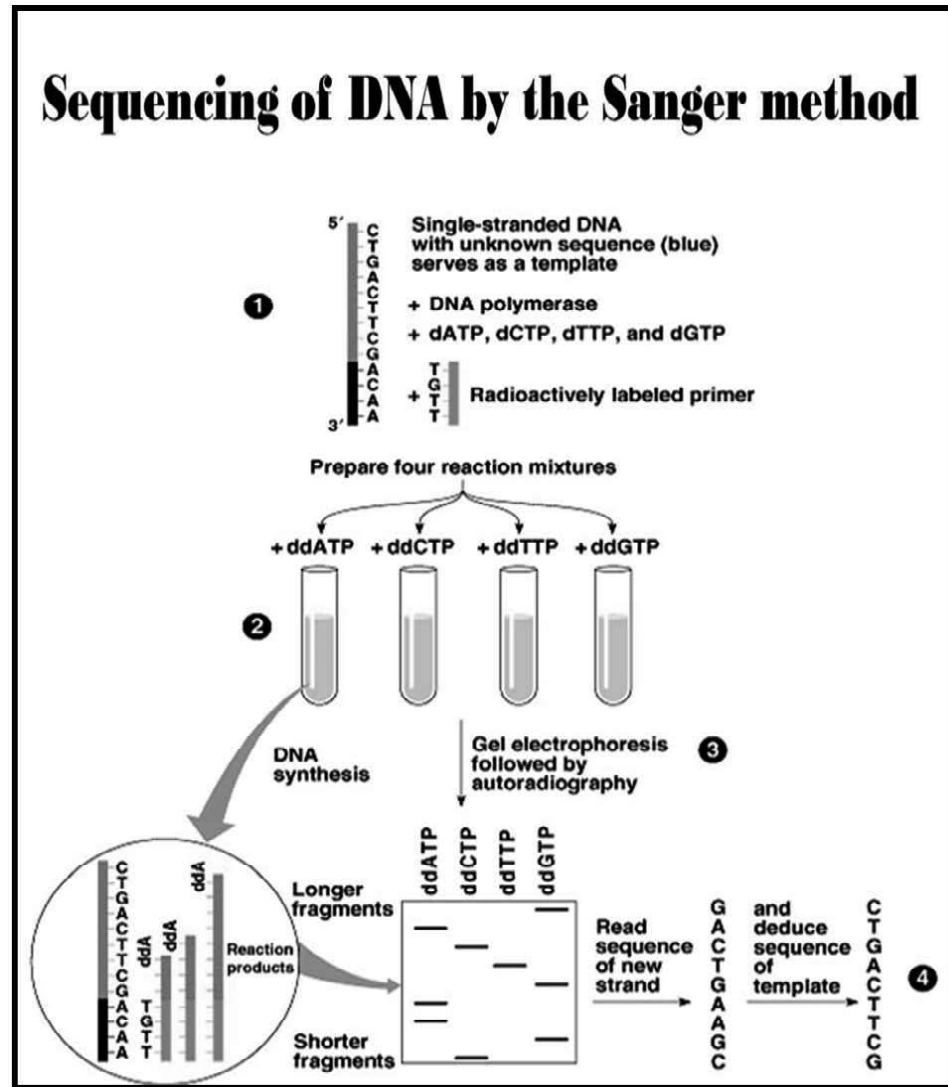


Fig. 2.25 Overview of Sanger Sequencing

Figure 2.25 is depicting the overview of Sanger sequencing.

Next-generation sequencing (NGS) is a second-generation sequencing method. Furthermore, it is a high-throughput DNA sequencing approach with the concept of massively parallel processing. Genome Analyzer/HiSeq/MiSeq (Illumina, Solexa), SOLiD System (Thermo Fisher Scientific), Ion PGM/Ion Proton (Thermo Fisher Scientific), and HeliScope Sequencer (Helicos BioSciences) are the numerous platforms presently performing next-

generation sequencing. Usually, they can sequence approximately 1 million to 43 billion short reads (50–400 bases each) per instrument run. Furthermore, the primary characteristic of next-generation sequencing is that it can accomplish a parallel examination of numerous targets. NGS has tremendously increased the speed as well as efficiency of detection of mutation. Mostly, in somatic cancer mutations, tumors are heterogeneous and comprises of both cancer cells as well as the normal cells. Though, the preparation of a DNA library by clonal amplification in next-generation sequencing for parallel sequencing helps to physically separate signals originating from each target DNA molecule in the library. Consequently, this allows the separation of DNA sequences of cancer cells from the DNA sequences of normal cells. Thus, all in all, the next-generation sequencing is a digital sequencing method with a higher depth of coverage variants.

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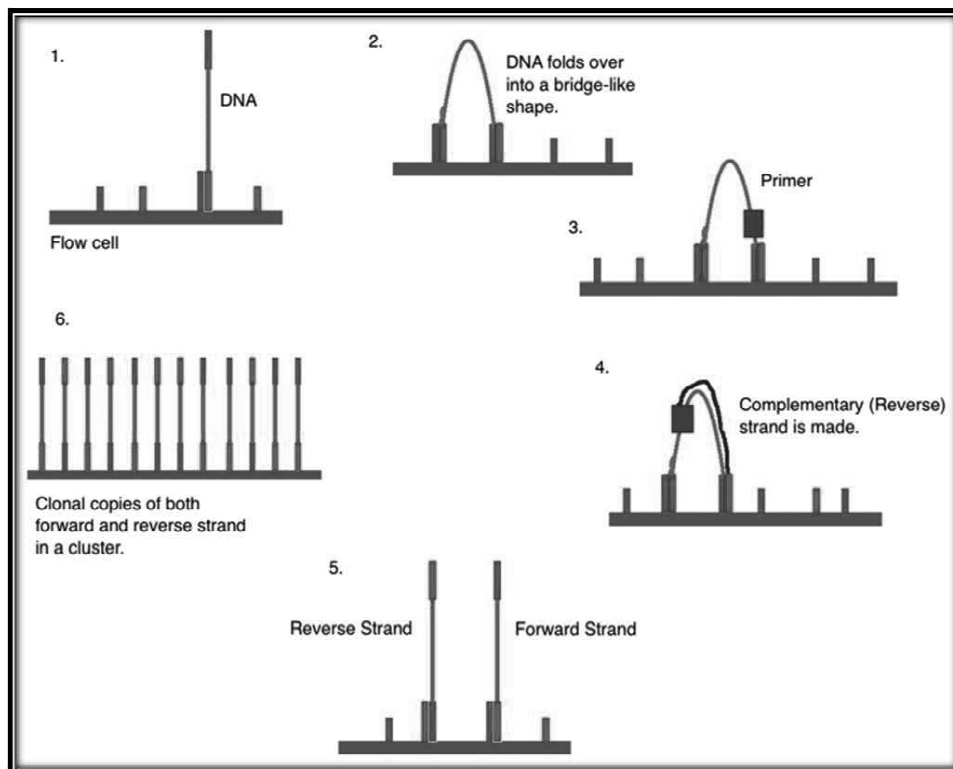


Fig. 2.26 Clonal Amplification in Illumine Sequencing

Figure 2.26 is depicting the clonal amplification in illumine sequencing.

(5) FISH Methods (Fluorescence in Situ Hybridization (FISH))

FISH is a special technique that can be used to view changes. In this methodology, a gene segment can be made to “light up” or fluoresce when it is bound to a special probe. The FISH technique is dependent upon hybridizing a probe with a fluorescent tag, which is complementary in sequence, to a short section of DNA on a target gene. The tag as well as the probe are applied to a sample of interest under conditions that enables the probe to attach itself to the complementary sequence in the specimen if it is present at all. After the specimen has been treated, excess of the fluorophore has been washed away and the sample can be visualized under a fluorescent microscope. By quantifying the amount of fluorescence with the help

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of a microscope, a skilled researcher or technician can easily determine the type of cell the probe was designed for is present, and if so, how much of it is present in a sample. Transcripts (mRNA) in microbes can also be targeted to detect if a specific gene is being expressed under the given conditions.

(6) Membrane filtration

A typical MF method for water analysis is performed by passing a known volume of water through a sterile membrane filter with a pore size small enough to retain bacterial cells (typically $0.45\mu\text{m}$). The filter is then transferred aseptically to the surface of an agar plate, or an absorbent pad saturated with a suitable selective medium and incubated. Colonies are allowed to develop on the surface of the filter and can be counted and examined directly. MF methods are quick and easy to perform, require little incubator space, and can handle large volumes of water if needed. Over the last 30 years, they have become the preferred methods for the microbiological examination of water for indicator organisms. There are several official published methods based on MF, notably a series of ISO methods, such as ISO 9308-1 for coliforms and *E. coli* and ISO 7899-2 for enterococci.

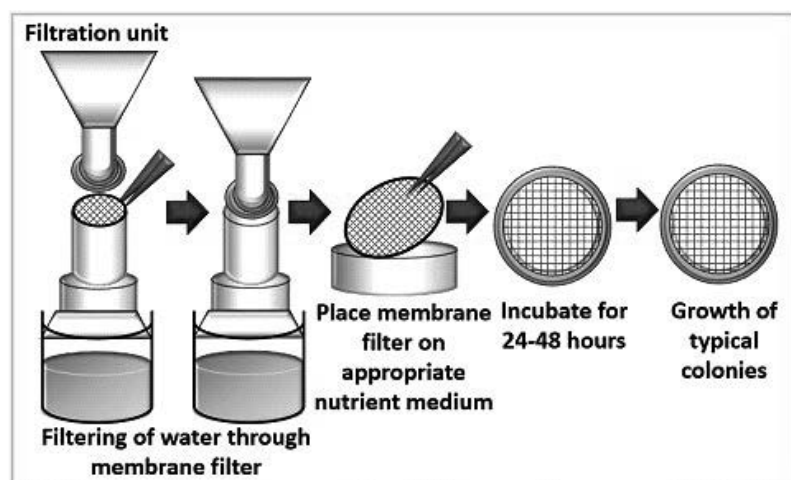


Fig. 2.27 Method of Membrane Filtration in Water Testing

Figure 2.27 is depicting the method of membrane filtration in water testing.

(7) Measuring Dissolved Oxygen present in the water

Dissolved oxygen is the amount of oxygen that is present in water. Water bodies receive oxygen from the atmosphere and from aquatic plants. Running water, such as that of a swift moving stream, dissolves more oxygen than the still water of a pond or lake. As mentioned above, oxygen gas is absorbed by the water from the atmosphere but it is being consumed by organic matter present in it. Therefore, if the oxygen gas present in water is than its normal saturation level, it indicates the presence of organic matter which makes the water unsuitable for consumption.

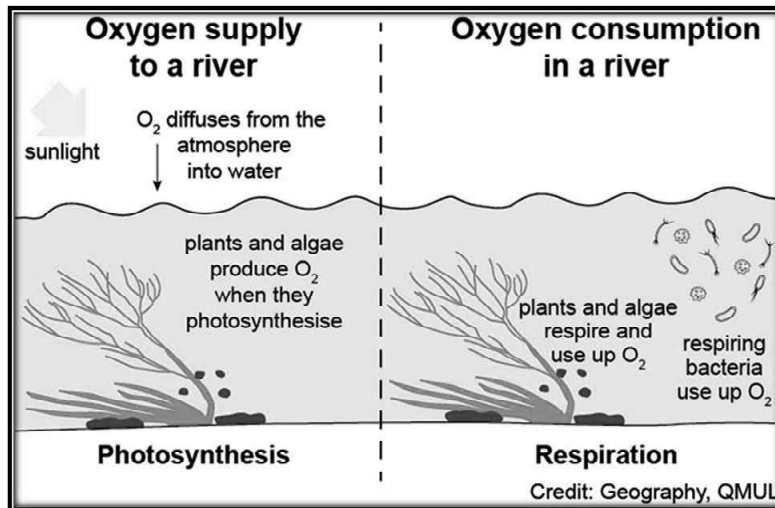


Fig. 2.28 Oxygen Supply and Consumption in a River System

Figure 2.28 is depicting the oxygen supply and consumption in a river system.

(8) Biological Oxygen Demand (BOD)

The amount of organic matter present in the water can be assessed by supplying oxygen to the water sample under observation. The organic matter present in water will start consuming the oxygen and undergoes oxidation. Thus, Biological oxygen demand (BOD) or Biochemical Oxygen demand (BOD) is referred as the amount of oxygen needed or demanded by aerobic microorganisms to break down the organic matter present in a certain sample of water at a specific temperature over a given period of time. BOD of water sample is evaluated by measuring the oxygen consumed by the bacteria from the decomposition of organic matter over a period of five days at a specific temperature of 20 degree Celsius. A lot of factors influence the BOD of water like temperature, pH of the water sample, inorganic substance present in the water, kind of aerobic microorganisms present in water, amount and type of organic matter present in the water sample etc. BOD values indicate the polluting potential of water. A BOD value of less than 1 mg/L indicates pure drinking water, BOD value between 2-8mg/L indicates moderately polluted water whereas a BOD value greater than 8mg/L indicates severely polluted water unfit for consumption. BOD finds its prime significance in sewage treatment plants. It indicates the respiration rate in sludge, soil, garbage etc.

Table 2.1 BOD Level and Corresponding Water Quality

| BOD Level (in ppm) | Water Quality |
|------------------------------|--|
| 1 - 2 | Very Good There will not be much organic waste present in the water supply. |
| 3 - 5 | Fair: Moderately Clean |
| 6 - 9 | Poor: Somewhat Polluted Usually indicates organic matter is present and bacteria are decomposing this waste. |
| 100 or greater | Very Poor: Very Polluted Contains organic waste. |

Table 2.1 is depicting the BOD level and corresponding water quality

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(9) Chemical Oxygen Demand (COD)

The chemical oxygen demand (COD) represents the amount of oxygen necessary for the aerobic biological oxidation (breakdown) of the organic substances in a given water body or sample to carbon dioxide (CO_2) and water (H_2O) if the organics are actually biodegradable. COD measurements are important in accessing the quality of a given water body. COD values help us to determine the extent of pollution that the water has undergone. COD tests also assess the amount of inorganic contaminants that is dissolved or suspended in a water body. Thus, it can be concluded that higher the COD, higher is the contamination level of water. The COD test usually involves a very strong oxidizing chemical agent like potassium dichromate ($\text{Cr}_2\text{O}_7^{2-}$). The primary basis for the COD test is that nearly all organic compounds can be fully oxidized to carbon dioxide with a strong oxidizing agent (e.g. $\text{Cr}_2\text{O}_7^{2-}$) under acidic conditions. Silver compounds can also be involved in the COD test to speed up the oxidation of certain organic compounds. Mercury can also be used in the COD test to reduce the interference from oxidation of chloride ions.

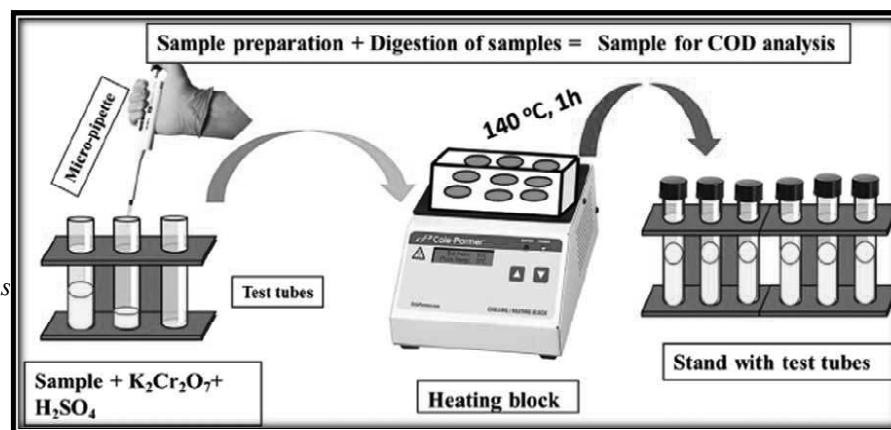


Fig. 2.29 COD Test

Figure 2.29 is depicting the COD test.

(10) Turbidity of water

One popular test measures the turbidity of the water. Turbidity refers to the measure of relative clarity of a liquid. It is an optical characteristic of water and is a measurement of the amount of light that is scattered by material present in the water when a light is passed through the water sample. Turbidity is directly proportional to light scattered i.e. higher the intensity of scattered light, higher is the turbidity of water. The material that increases the turbidity of water includes silt, tiny organic matter, inorganic material, clay, plankton, dissolved matter as well as other microscopic organisms. Higher turbidity makes the water cloudy or opaque. In fact, turbidity of the water is an indicator of potential pollution present in the water bodies. Excessive turbidity, cloudiness or opaqueness in drinking water is not only unappealing, but also unhealthy. High turbidity can provide habitat for pathogens which may lead to outbreak of water borne diseases. As mentioned above, turbidity of water directly influences the penetration of light. It limits the penetration of light in the aquatic bodies. Productivity of the fresh water system

depends upon the penetration of the light. For instance: Large shallow lakes tend to be more productive when compared to lakes with deep water as more light reaches the lake bed. In larger rivers, the canopy is open which allows the algal production. The turbidity of the water is measured by using an instrument known as turbidity rod or by a turbidity meter with optical observations. Turbidity rod consists of a graduated aluminium rod to give turbidity directly in silica units (mg/l). Turbidity meter works on the principle of measuring the interference caused by the water sample to the passage of light rays. Turbidity of water is expressed as the amount of suspended matter in mg/l or parts per million (ppm). The standard unit is that which is produced by one milligram of finely divided silica (fuller's earth) in one litre of distilled water.

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Check Your Progress

11. What is FISH method?
12. Define membrane filtration (MF) method?
13. What is BOD stands for?

2.6 SEWAGE WASTE WATER TREATMENT

Sewage is created by residential, institutional, commercial as well as industrial establishments which includes household waste liquid from toilets, baths, showers, kitchens, sinks and so forth that is disposed of via sewers. In several areas, sewage also includes liquid waste from industry. Sewage can be effectively treated at the point where it is created like in septic tanks or it can be collected and transported via a network of pipes and pump stations to a municipal treatment plant. For managing/controlling water pollution from point sources (like sewage), treatment of waste water is essential before being discharged into aquatic bodies. Parameters which needs to be taken into consideration before discharging water are Total solids, Biological Oxygen Demand (BOD), Chemical Oxygen Demand (COD), nitrates and phosphates, oil and grease, toxic metals etc. Waste waters should be properly treated by primary, secondary and tertiary treatments to reduce the BOD, COD levels up to the permissible levels before discharging them into aquatic bodies.

Step-by-Step- Waste Water Treatment Process

Sewage treatment, or domestic wastewater treatment refers to the process of removing physical, chemical and biological contaminants/pollutants from them. The main objective of the waste water treatment is to produce an environmentally-safe fluid waste stream (or treated effluent) or reuse the water either by humans/animals or for any other purpose like as farm fertilizer. Conventional sewage treatment involve three stages namely primary, secondary and tertiary treatment. Step by step process of waste water treatment is as follow:

Step One: Collection of Wastewater

- Collection of wastewater is the first step in the process of waste water treatment.

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- The collection system is set in place by municipal administration as well as the private business owners.
- These collection centres ensure that all the wastewater is collected together at one place and then is directed into the central point.
- Once collected, wastewater is directed into the treatment plant by using the underground drainage systems.
- Apart from the underground drainage system, Exhauster tracks can also be used to direct the collected wastewater into the treatment plant.
- All the precautions like wearing protective clothing and gloves should be taken before transporting this collected wastewater into the treatment plant. Transportation should be done in hygienic conditions.
- It is also essential to ensure that the pipes or tracks do not leak

Pre-Treatment

- Pre-treatment involves removing large solid items like sanitary items, diapers, wipes, broken bottles etc. or debris from the raw sewage. This is an essential step as allowing such items further in the process can damage the equipment used for treating waste water.
- Grit and sand are also removed in the process.
- After this, it is rinsed in the sewer.
- All the debris are then dried up and incinerated.
- Sand and grit are then settled out in the tanks.

Odor Control

- Wastewater also contains several dirty substances that may give it foul smell.
- Odor treatment is mainly initiated at the treatment plant.
- Odor treatment is done by containing the wastewater and treating it with chemicals.
- This chemical will automatically neutralize the odor producing elements in the wastewater.
- Odor control/treatment helps in keeping the environment and surroundings smell free.

Screening

- Screening involves removing all the large items/objects that may have remained even after the initial step of pre-treatment.
- Screening is done more rigorously to ensure all solid objects are removed from the waste water.
- A rigorous screening ensures clearing all the solid objects that may potentially damage the equipment later on.
- Screening also helps in removing all the unwanted solid material from waste water.
- All these solid wastes are transported and disposed off in the landfills.

Step Two: Primary Sewage Treatment

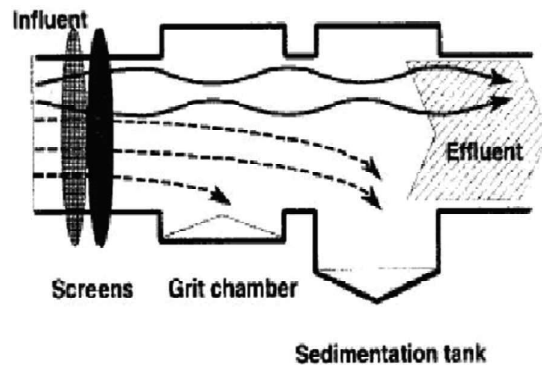


Fig. 2.30 Process of Primary Treatment

Figure depicting the process of primary treatment.

- Primary sewage treatment involves separation of solid macrobiotic matter from wastewater.
- This process is done by pouring wastewater into the big tanks (known as big/large settlement tanks) leading to the settlement of the solid matter on the tank's surface.
- At this stage, waste water gets separated into two: first is the solid organic matter settled at the bottom of the tank and second is the remaining liquid.
- The denser waste that sinks at the bottom of the settlement tank is known as sludge.
- The wastes are then pushed to the center by large scrappers which continuously remove them from the bottom.
- At the top, there is another layer of fats which floats since it is less dense than water. This layer of fats forms a crust.
- The water between the crust and the sludge, which is now homogenous, is taken to for secondary treatment.
- This makes the treatment more proficient.

Step Three: Secondary Sewage Treatment

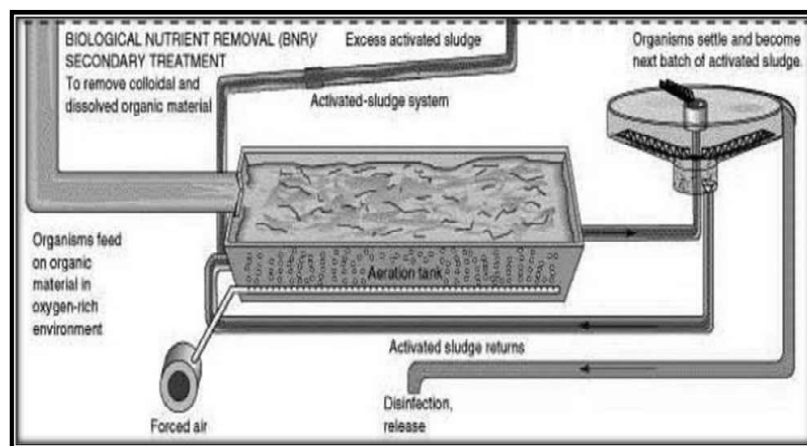


Fig. 2.31 Process of Secondary Sewage Treatment

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- Figure 2.31 depicting the process of secondary sewage treatment a
- Secondary treatment is also known as activated sludge process
 - At this stage, Oxygen (O_2) is introduced into the water.
 - This helps break down the sludge that may have escaped the primary treatment.
 - It begins with pumping air into the huge aeration tanks that has a mix of wastewater with a seed sludge.
 - A small amount is pumped in to activate the growth of aerobic bacteria (which uses oxygen).
 - This step also activates the growth of microorganisms which consume all the remaining organic matter.
 - After sometime, debris produced will settle down at the bottom of the large tank.
 - Water can pass through the huge tank in three to six hours
 - Humus and the dead bacteria settle out in another chamber for extra treatment.

Bio-Solids Handling

- The ultimate solid matter that settles down after the primary and secondary treatment procedures, are then directed into the digesters.
- Digesters are then heated strongly at the room temperature.
- Solid matter present inside the digesters undergoes anaerobic digestion.
- Production of methane gas and the formation of nutrient rich bio-solids are observed at this stage
- Recycled methane gas is mostly used in the local firms as primary source of energy
- It can also be used as the energy source in waste water treatment plant (for running equipment used during the process)

Step Four: Tertiary Treatment

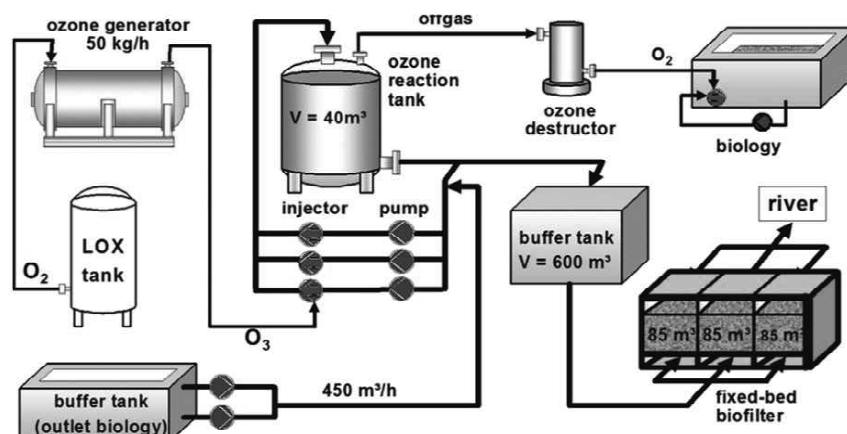


Fig. 2.32 process of tertiary treatment

Figure 2.32 depicting the process of tertiary treatment in waste water treatment plant.

- Tertiary treatment is employed where the discharge points is very sensitive/ ecologically important
- These sensitive points include streams or dams where the water will be used directly by plants/animals/people.

Tertiary treatment involves:

- a. Chlorination of waste water
 - b. Ozone treatment of waste water
 - c. Use of Ultra Violet light to disinfect waste water
 - d. Reducing the level of Nitrogen in waste water
- At tertiary treatment stage, all the remaining impurities are removed from the wastewater.
 - Tertiary treatment stage is almost similar to that used in the drinking water treatment plant.
 - It helps in the removal of 99% impurities in waste water.
 - At this stage, wastewater is treated so rigorously that it produces effluent water having almost same quality as that of the drinking water.
 - Tertiary treatment of waste water is highly expensive and time-consuming process
 - It also requires sophisticated equipment, strong chemicals, constant supply of energy as well as skilled labour.

Disinfection

- Disinfection is an integral process of waste water treatment plan
- Even after going through such rigorous processes, some diseases causing organisms might be left in the water.
- In order to eradicate disease causing microorganisms, wastewater is disinfected with the mixture of sodium hypochlorite and chlorine for about 20-25 minutes

Step Five: Sludge Treatment

- Sludge produced and collected at the primary and secondary treatment levels of the waste water treatment plans requires a very high concentration and thickening process.
- This can be achieved by putting them into the thickening tanks to settle down and then separate it later from the water.
- The entire process of sludge treatment may take up to 24 hours.
- After sludge treatment, the remaining water will then be collected and reversed back to the large aeration tank to enable further treatment.
- The treated water is recycled for agricultural purposes
- It can also be discharged into lakes/streams/rivers.

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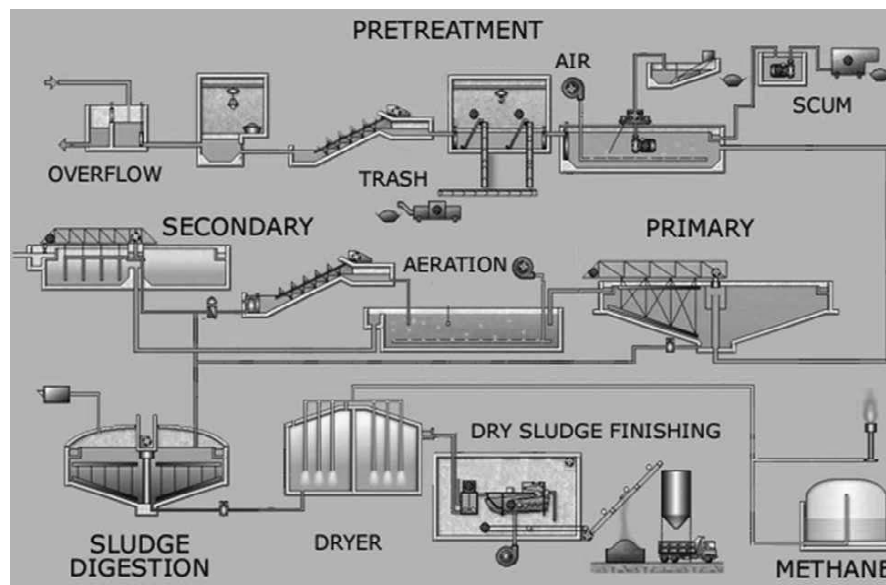


Fig. 2.33 Entire Process of Waste Water Treatment

Figure 2.33 depicts the entire process of waste water treatment.

Methods of waste water treatment

There are three methods of waste water treatment namely:

- (1) Biological waste water treatment
- (2) Physical waste water treatment
- (3) Chemical waste water treatment

(1) Biological Wastewater Treatment:

- Biological treatment of wastewater is done to obtain water fit for human consumption.
- Biological treatment involves the extensive use of micro- bacteria that decompose the waste material or organic impurities and improve the quality of water to be used for households.

Three common methods of biological treatment are as follows: -

- (a) **Aerobic Process:** As the name suggests, oxygen is required for this purpose. During the aerobic process, oxygen consuming bacteria decompose and devour the organic impurities of wastewater and convert it to CO₂ for reproduction and growth.
- (b) **Anaerobic Process:** This process occurs in the absence of oxygen. In the Anaerobic or fermentation process, waste or sludge present in the wastewater is fermented at a specified temperature.
- (c) **Composting:** Composting is a kind of Aerobic process, in which sludge or biosolid is mixed with carbon sources in the presence of oxygen to remove all the impurities.

(2) Physical Wastewater Treatment:

- Physical wastewater treatment involves the use of physical approaches to remove impurities.

Three important physical methods strategies used to clean waste water:-

- (a) **Sedimentation:** Sedimentation refers to the process of removing insoluble debris from the water. The first unit of sedimentation tank used during the process are known as “primary sedimentation tank” or “primary settling tanks”. Solids or dense materials like stones, dust particles, debris are removed by gravity force, and other less dense particles are carried to the next tank.
- (b) **Aeration:** In this method of water treatment, the air is made to circulate through the water to give it oxygen to increase its Bio-Oxygen Demand.
- (c) **Filtration:** Filtration is used for filtering out impurities present in wastewater. One of the most common filter used is Sand Filter.

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(3) Chemical Wastewater Treatment:

- Chemical wastewater treatment involves the use of different strong chemicals to clean water.
- Chlorination- using chlorine (that acts as potent oxidising chemical) for treating the waste water is done to kill harmful bacteria.
- Ozone is also an oxidizing disinfectant that works to destroy viruses and harmful bacteria in the water.
- Neutralization method is used for treating industrial waste water. In this method, base or acid is mixed with water till it reaches the pH value of water. Generally, lime is used as a base to neutralize acidic water.

Advantages of waste water treatment

Wastewater treatment has a number of benefits.

- Wastewater treatment ensures the release of pollutant/contaminant free water into lakes/streams/rivers.
- Waste water treatment helps in reducing the water pollution at surface level
- Waste water treatment prevents the outbreak of water borne diseases.
- Water obtained from waste water treatment plants can be used for multiple purposes like human consumption/agricultural purposes/industrial use etc. This helps in the effective management of an essential natural resource, i.e., water.
- A lot of by-products released during treating water like methane gas can be used as source of energy.
- Recovered metal particles from the waste water treatment plant can be used effectively.

Check Your Progress

14. What is sewage treatment, or domestic wastewater treatment?
15. Which are the three methods of waste water treatment?
16. Define composting.
17. What is physical wastewater treatment?

2.7 ANSWERS TO 'CHECK YOUR PROGRESS'

NOTES

1. A biogeochemical cycle is referred to as the pathway via which a chemical substance is turned over or moves through the biotic (biosphere) and the abiotic (lithosphere, atmosphere and hydrosphere) compartments of Earth.
2. Essential nutrients added to the ecosystem by rock weathering process are Calcium (Ca), magnesium (Mg), potassium (K), sodium (Na), silicon (Si), iron (Fe), aluminum (Al), and phosphorus (P).
3. By process of respiration and decomposition of dead organic matter, carbon returns back to the atmosphere.
4. Nitrogen fixation refers to the conversion of N_2 to ammonia or related nitrogenous compounds by Blue green algae (Cyanobacteria) like Anabaena, Nostoc, Azolla, etc.
5. Silica enters into the aquatic environments by weathering of rock-forming silicate minerals by acidic dissolution.
6. Many microorganisms play foundational roles in aquatic ecosystems, capturing the sun's energy through photosynthesis and, through their role in decomposition, releasing nutrients stored in organic tissue.
7. Archaeobacteria are the most ancient bacteria found in the most extreme habitats such as salty area (halophiles), hot springs (thermoacidophiles) as well as marshy areas (methanogens).
8. Nostoc, Anabaena, Spirulina are some of the best examples of cyanobacteria.
9. Phytoplankton are small microscopic, unicellular and photosynthetic organisms which freely float in water bodies.
10. The region of a water body near the shoreline is known as littoral zone.
11. FISH is a special technique that can be used to view changes. In this methodology, a gene segment can be made to "light up" or fluoresce when it is bound to a special probe.
12. A typical MF method for water analysis is performed by passing a known volume of water through a sterile membrane filter with a pore size small enough to retain bacterial cells (typically $0.45\mu\text{m}$).
13. Biological oxygen demand (BOD) or Biochemical Oxygen demand (BOD) is referred as the amount of oxygen needed or demanded by aerobic microorganisms to break down the organic matter present in a certain sample of water at a specific temperature over a given period of time.
14. Sewage treatment, or domestic wastewater treatment refers to the process of removing physical, chemical and biological contaminants/pollutants from them.
15. There are three methods of waste water treatment namely
 - Biological waste water treatment
 - Physical waste water treatment
 - Chemical waste water treatment

16. Composting is a kind of Aerobic process, in which sludge or bio-solid is mixed with carbon sources in the presence of oxygen to remove all the impurities.
17. Physical wastewater treatment involves the use of physical approaches to remove impurities

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2.8 SUMMARY

- A biogeochemical cycle is referred to as the pathway via which a chemical substance is turned over or moves through the biotic (biosphere) and the abiotic (lithosphere, atmosphere and hydrosphere) compartments of Earth.
- Energy flows via an ecosystem and is released as heat, however chemical elements are recycled.
- In the gaseous cycle, the main reservoir of nutrients is either the atmosphere or the hydrosphere (ocean).
- In the sedimentary cycle, the main reservoir is the lithosphere and the sedimentary as well as other rocks of the earth's crust.
- In the Closed system, chemicals or elements used in the ecosystem are recycled instead of being lost.
- In the open system, chemicals or elements tend to flow in and out of the system.
- Carbon dioxide is the basic building block that most autotrophs use to build multi-carbon, high-energy compounds like glucose.
- Carbon dioxide is the element which anchors all organic substances from coal and oil to DNA (deoxyribonucleic acid: the compound that carries genetic information).
- Nitrogen is vital for several processes and is crucial for life on Earth. The nitrogen cycle is the biogeochemical cycle by which nitrogen is converted into multiple chemical forms as it circulates among atmosphere, hydrosphere and lithosphere.
- Plants take in the nitrogen compounds which are available in the form of ammonia, nitrite ions, nitrate ions or ammonium ions and are used in the formation of the plant as well as animal proteins.
- Phosphorous is an essential component of biomolecules like ADP and ATP, nucleic acids, phospholipids (membranes), apatite (bones and teeth), etc.
- Sulphur is a yellow, brittle, tasteless, odorless non-metal. It is one of the most abundant elements in nature and is present in almost all kinds of proteins.
- The sulphur is released by geochemical and meteorologic processes like the weathering of rocks.
- Iron is an essential micronutrient for almost every life form present on earth.
- The iron cycle influences other cycles like nitrogen, phosphorous as well as sulphur cycles.

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- There are bacteria that oxidise manganese to insoluble oxides, and others that reduce it to Mn^{2+} in order to use it.
- Most protozoal species move independently by one of three types of locomotor organelles: flagella, cilia, and pseudopodia.
- Most protozoans have a single nucleus, but some have both a macronucleus and a micronuclei.
- Phytoplankton composed of both eukaryotic as well as prokaryotic species which colonizes upper euphotic part of the water column ranging from freshwater to ocean conditions.
- The name zooplankton is derived from the Greek: Zoon, animal; planktos, wandering.
- The region of a water body near the shoreline is known as littoral zone.
- Algae and Diatoms are the most common phytoplankton present in potamon zone of streams.
- One of the most effective way to check water supplies for faecal contamination is microbiological analysis, and a range of test methods designed for that purpose has been developed for the water industry.
- For several years, the primary method of choice was the multiple tube 'Most Probable Number' (MPN) technique.
- A typical MF method for water analysis is performed by passing a known volume of water through a sterile membrane filter with a pore size small enough to retain bacterial cells (typically $0.45\mu m$).
- Biological oxygen demand (BOD) or Biochemical Oxygen demand (BOD) is referred as the amount of oxygen needed or demanded by aerobic microorganisms to break down the organic matter present in a certain sample of water at a specific temperature over a given period of time.
- Sewage treatment, or domestic wastewater treatment refers to the process of removing physical, chemical and biological contaminants/pollutants from them.
- Biological treatment of wastewater is done to obtain water fit for human consumption.
- Physical wastewater treatment involves the use of physical approaches to remove impurities.

2.9 KEY TERMS

- **Biogeochemical Cycle:** A biogeochemical cycle is referred to as the pathway via which a chemical substance is turned over or moves through the biotic (biosphere) and the abiotic (lithosphere, atmosphere and hydrosphere) compartments of Earth.
- **Leaching:** Leaching occurs when water flowing vertically through the soil transports nutrients in solution downward in the soil profile. The process can leads to loss of essential nutrients.

- **Nitrogen fixation:** Nitrogen fixation refers to the conversion of N_2 to ammonia or related nitrogenous compounds by Blue green algae (Cyanobacteria) like Anabaena, Nostoc, Azolla, etc.
- **Iron cycle:** The iron cycle (Fe) is the biogeochemical cycle of iron via the atmosphere, hydrosphere, biosphere and lithosphere.
- **Trophozoite:** A free-living vegetative form of a protozoa is known as trophozoite.
- **Phytoplankton:** Phytoplankton are small microscopic, unicellular and photosynthetic organisms which freely float in water bodies.
- **Zooplankters;** Zooplankters are microscopic, unicellular or multicellular forms with size ranging from a few microns to a millimeter or even more.
- **Littoral zone:** The region of a water body near the shoreline is known as littoral zone.
- **Membrane filtration method:** A typical MF method for water analysis is performed by passing a known volume of water through a sterile membrane filter with a pore size small enough to retain bacterial cells (typically $0.45\mu m$).
- **Dissolved oxygen:** Dissolved oxygen is the amount of oxygen that is present in water.
- **Biological oxygen demand:** Biological Oxygen Demand (BOD) is referred as the amount of oxygen needed or demanded by aerobic microorganisms to break down the organic matter present in a certain sample of water at a specific temperature over a given period of time.
- **Turbidity:** Turbidity refers to the measure of relative clarity of a liquid.
- **Sewage treatment:** Sewage treatment or domestic wastewater treatment refers to the process of removing physical, chemical and biological contaminants/pollutants from them.
- **Composting:** Composting is a kind of Aerobic process, in which sludge or biosolid is mixed with carbon sources in the presence of oxygen to remove all the impurities.
- **Physical wastewater treatment:** Physical wastewater treatment involves the use of physical approaches to remove impurities.

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2.10 SELF-ASSESSMENT QUESTIONS AND EXERCISES

Short-Answer Questions

1. How nutrients does enters into the ecosystem?
2. In which form carbon is present in the atmosphere?
3. Why Iron is considered as an essential micronutrient for almost every life form present on earth?
4. How does the iron cycle influences other cycles?

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5. State the characteristics of bacteria.
6. Why the Fungi is considered both beneficial as well as harmful?
7. Which are the chief primary producers in lentic water system?
8. In which areas water-borne diseases are more likely widespread?
9. Why the water borne diseases are regarded to be important for public health?
10. What do you understand by turbidity of water?
11. State the advantages of waste water treatment.

Long-Answer Questions

1. Briefly explain the flow of nutrients in an ecosystem.
2. Describe various phases of nitrogen cycling in aquatic Ecosystem.
3. Explain nitrogen cycle in detail with diagram.
4. Describe the freshwater micro-biota in detail.
5. Write a short note on sea water microbiota.
6. Explain the Membrane Filtration (MF) method with diagram.
7. Describe step-by-step- waste water treatment process. in detail.

2.11 FURTHER READING

- Atlas, Ronald M., Richard Bartha. 1993. *Microbial Ecology Fundamentals and Applications*. U.S.: Benjamin/Cummings Publishing Company.
- Chénard, Caroline, Federico M. Lauro. 2017. *Microbial Ecology of Extreme Environments*. U.S.: Springer International Publishing.
- Alexander, Martin. 1991. *Introduction to Soil Microbiology*. U.S.: R.E. Krieger Publishing Company.
- David, L. Kirchman. 2012. *Processes in microbial ecology*. Oxford: Oxford University Press.

UNIT 3 BIOLOGY OF PARASITISM I

Structure

- 3.0 Introduction
- 3.1 Objectives
- 3.2 General Introduction and Terms Employed
 - 3.2.1 Classes of Parasites and Host
 - 3.2.2 Spread of Parasitic Infection and Host Parasite Relationship
- 3.3 Protozoan Parasites Pathogenic to Man
- 3.4 Geographical Distribution, Habitat, Morphology, Lifecycle, Pathogenesis, Clinical Features, Lab Diagnosis of some Protozoan Parasites
 - 3.4.1 Protozoan Parasites belong to Sporozoa and Ciliata
- 3.5 Answers to 'Check Your Progress'
- 3.6 Summary
- 3.7 Key Terms
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- 3.9 Further Reading

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3.0 INTRODUCTION

Parasitism is a close relationship between species, where one organism, the parasite, lives on or inside another organism, the host, causing it some harm, and is adapted structurally to this way of life. The entomologist E. O. Wilson has characterized parasites as “predators that eat prey in units of less than one”. Parasitism occurs when one organism (the parasite) takes nutrients from another (the host). The host is usually weakened by the parasite as it siphons resources the host would normally use to maintain itself. Parasites do not necessarily kill their hosts. When they do, it is often a slow process, allowing the parasite time to complete its reproductive cycle before it or its offspring are able to spread to another host. A parasite may remain attached to the same host for its full lifespan, but some parasites have complex life cycles involving multiple host species. Parasites infect many types of organisms, including other animals and plants. For example, fleas and roundworms are common dog parasites. Plants can be infected by fungi, bacteria, and viruses; there are also plants that parasitize other plants. Even bacteria can be parasitized by viruses called bacteriophages. Like predation, parasitism is a type of consumer-resource interaction, but unlike predators, parasites, with the exception of parasitoids, are typically much smaller than their hosts, do not kill them, and often live in or on their hosts for an extended period. Parasites of animals are highly specialized, and reproduce at a faster rate than their hosts. Classic examples include interactions between vertebrate hosts and tapeworms, flukes, the malaria-causing *Plasmodium* species, and fleas

In this Unit you will study about general introduction of parasites and host, information for spread of parasitic infection, host parasite relationship, geographical distribution, habitat, morphology, lifecycle, pathogenesis, clinical features, lab diagnosis, treatment and prophylaxis of protozoan parasites pathogenic to man such as rhizopoda, flagellata, sporozoa ciliata.

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3.1 OBJECTIVES

After going through this unit you will be able to:

- Give general introduction of parasites and host
- Elaborate on the information for spread of parasitic infection
- Analyze host parasite relationship
- Explain morphology, lifecycle, pathogenesis, clinical features, lab diagnosis treatment and prophylaxis of protozoan parasites

3.2 GENERAL INTRODUCTION AND TERMS EMPLOYED

Parasitism is a close relationship between species, where one organism, the parasite, lives on or inside another organism, the host, causing it some harm, and is adapted structurally to this way of life. The word parasite is derived from the Latin form of the Greek word, meaning “one who eats at the table of another”. Parasitism is generally defined as a relationship between the two living species in which one organism is benefitted and cause harm to the other. The organism that is benefitted is called the parasite, while the one that is harmed is called the host. A few examples of parasites are tapeworms, fleas, and barnacles. Tapeworms are flatworms that are found attached to the insides of the intestines of animals such as cows, pigs, etc. They feed on the host’s partly digested food, depriving it of the nutrients. The entomologist E. O. Wilson has characterized parasites as “predators that eat prey in units of less than one”. Parasites include single-celled protozoans such as the agents of malaria, sleeping sickness, and amoebic dysentery; animals such as hookworms, lice, mosquitoes, and vampire bats; fungi such as honey fungus and the agents of ringworm; and plants such as mistletoe, dodder, and the broomrapes. There are six major parasitic strategies of exploitation of animal hosts, namely parasitic castration, directly transmitted parasitism (by contact), tropically transmitted parasitism (by being eaten), vector-transmitted parasitism, parasitoidism, and micropredation.

Parasitism is a kind of symbiosis, a close and persistent long-term biological interaction between a parasite and its host. Unlike saprotrophs, parasites feed on living hosts, though some parasitic fungi, for instance, may continue to feed on hosts they have killed. Unlike commensalism and mutualism, the parasitic relationship harms the host, either feeding on it or, as in the case of intestinal parasites, consuming some of its food. Because parasites interact with other species, they can readily act as vectors of pathogens, causing disease.

People have known about parasites such as roundworms and tapeworms since ancient Egypt, Greece, and Rome. In Early Modern times, Antonie van Leeuwenhoek observed *Giardia lamblia* in his microscope in 1681, while Francesco Redi described internal and external parasites including sheep liver fluke and ticks. Modern parasitology developed in the 19th century. In human culture, parasitism has negative connotations. Modern parasitology developed in the 19th century

with accurate observations by several researchers and clinicians. In 1828, James Annersley described amoebiasis, protozoal infections of the intestines and the liver, though the pathogen, *Entamoeba histolytica*, was not discovered until 1873 by Friedrich Lösch. James Paget discovered the intestinal nematode *Trichinella spiralis* in humans in 1835. James McConnell described the human liver fluke in 1875. A physician at the French naval hospital at Toulon, Louis Alexis Normand, in 1876 researching the ailments of French soldiers returning from what is now Vietnam, discovered the only known helminth that, without treatment, is capable of indefinitely reproducing within a host and causes the disease strongyloidiasis. Patrick Manson discovered the life cycle of elephantiasis, caused by nematode worms transmitted by mosquitoes, in 1877. Manson further predicted that the malaria parasite, *Plasmodium*, had a mosquito vector, and persuaded Ronald Ross to investigate. Ross confirmed that the prediction was correct in 1897–1898. At the same time, Giovanni Battista Grassi and others described the malaria parasite's life cycle stages in *Anopheles* mosquitoes.

Host parasite interactions are crucial for parasitology studies. The host gene array complements to the parasite gene array and studying the host cell undergoing infection, transcription profiling, growth latency, and mortality gives a clear idea about the development and survival of parasites in the host cell.

The following are some of the definitions of the terms associated with host–parasite interaction:

- **Parasites:** These are living organisms that live in or on a host, which provides physical protection and nourishment.
- **Medical parasitology:** It deals with parasites, which infect man and the diseases produced by them.
- **Host:** It is an organism, which harbours the parasites. The host offers some resistance to the injury done by the parasite and there may be some adaptation or tolerance between the parasite and the host.
- **Symbiosis:** It is an association in which both parasite and host are so dependent upon each other that they cannot live without the help of the other, and both the partners do not face any harm from the association.
- **Commensalism:** It is an association in which the parasite derives benefit without causing any harm to its host.
- **Parasitism:** It is an association of host and parasite in which that latter is the beneficiary and the former gets nothing in return, and on the contrary, suffers some injury. The degree of dependence of the parasite on the host is variable and is different in different organisms.
- **Free-living parasite:** It is a parasitic stage of active existence, in which the parasite lives independently of a host.
- **Zoonosis:** It is a parasitic infection that is generally and primarily confined to vertebrate animals, but can sometimes produce disease in humans if they become infected accidentally. These infections are called zoonotic infections or zoonosis; for example, leishmaniasis, African trypanosomiasis, South American trypanosomiasis, japonicum schistosomiasis, trichinellosis and echinococcosis.

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- Anthroponoses: It is a parasitic infection that is confined to humans, for example, malaria and filaria.

The parasite is usually much smaller than the parasite host. For example, the intestine of some vertebrates serves as a host for the parasite called tapeworms. This parasite remains small enough to fit in the host's intestinal tract although it has a capacity to grow long.

A roundworm or *Trichinella spiralis* causes a disease called trichinosis. In this case, the host has both reproductive adults in its digestive tract and immature juveniles in its muscles. Therefore, the host is recognized as both intermediate host and a definitive host.

Parasitic bacteria: These are the heterotrophic bacteria, characterized by acquiring their nutrition from other organisms. The parasitic bacteria require a host to survive and such a process may either cause harm to the host or benefit it.

Types of Parasitism

Depending on host–parasite interaction, you can classify the parasitism in two types – temporary parasitism and permanent parasitism. Temporary parasitism is also called partial or intermittent parasitism.

3.2.1 Classes of Parasites and Host

Knowledge about the classes and life cycles of parasites is important in their detection and in treating the diseases caused by them.

Classes of Parasites

There are two classes of parasites as follows:

- (i) Ectoparasites:** These parasites inhabit the surface of the host's body without penetrating into the tissues. They are ticks and fleas and called ectoparasites. They cause an infestation and are important as vectors transmitting pathogenic micro-organisms.
- (ii) Endoparasites:** These parasites live within the host's body in the blood, tissues, body cavities, digestive tract and other organs. Endoparasites cause an infection. Most of the parasites causing human disease are endoparasites.

Life Cycles of Parasites

Parasites can pass through a series of developmental stages. Finally, a stage is attained when the organism reproduces and a new cycle of development begins. Parasites' life cycles differ in different species and they may be either sexual or asexual though in certain parasites, the sexual multiplication is immediately followed by asexual multiplication.

Broadly, there are two types of life cycles of parasites, which are as follows:

- (i) Direct life cycle:** When a parasite requires just one species of host to complete its development, it is called the direct life cycle, e.g., *Entamoeba histolytica* (it causes amoebiasis in man) requires only a single human host for the completion of its life cycle.
- (ii) Indirect life cycle:** It is a stage in which a parasite requires two or more species of hosts to complete its life cycle. For example, filarial

worms (they cause filariasis in man) require both a human host and an insect host (mosquito) in which they complete their development.

Types of hosts in cycle of parasites

The following types of hosts are seen in parasitology:

1. **Definitive host:** It is a kind of host in which the parasite spends the adult stage of its life or utilizes a sexual method of reproduction. In most human parasitic infections, man is the definitive host; however, man acts as intermediate host in malaria and hydatid disease.
2. **Intermediate host:** It is a kind of host in which the larval stage of the parasite either lives or the asexual multiplication takes place. Some times, larval developments are completed in two different intermediate hosts called first and second intermediate hosts respectively.
3. **Paratenic host:** It is kind of host in which a parasite merely remains viable without any further development. Such host can transmit the infection to another host. Such hosts are also called a transport hosts or mechanical hosts, e.g., a fly may transfer amoebic cysts from infected faeces to food that is eaten by humans.
4. **Reservoir host:** It is a kind of host that sustains a parasitic infection in an endemic area. A reservoir host is an animal or man in which a parasite commonly resides or one in which a parasite that infects man is able to be maintained in the absence of a human host.

In majority of parasitic infections, man is the main reservoir. In some cases, both man and animals are infected and the latter serves as reservoirs of infection, e.g., Rhodesian trypanosomiasis (antelope), Trichinellosis (pig and rat) and hydatid disease (sheep and cattle).

Mechanisms of Transmission of Parasitic Infections

Although the basic methods of transmission of parasitic infections have been discussed earlier in this unit, some of the more advanced methods are as follows:

- By ingestion through oral ways, through contaminated food or water, or from soiled fingers or fomites. This method of transmission is also known as the faecaloral route.
- By ingesting food or drinks contaminated by infective forms of parasites, e.g., cysts of *Entamoeba histolytica*, and the ova of *Ascaris iumbricoides*, *Trichuristrichuria* and *Enterobius vermiculares*.
- By ingesting an infective form of parasite in raw or undercooked meat of an intermediate host like beef containing the larval stages of *Taenia saginata* (*cysticercus bovis*) and the larval forms of *Trichinella spiralis*; pork containing the larval forms of *Taenia solium* (*cysticercus cellulose*); fish containing the plerocercoid larvae of *Diphyllobothrium latum*; and crab or crayfish containing the metacercarial stages of *Paragonimus westermani*.
- By drinking the parasite in water containing infected cyclops infected with the larval forms of *Dracunculus medinensis*.

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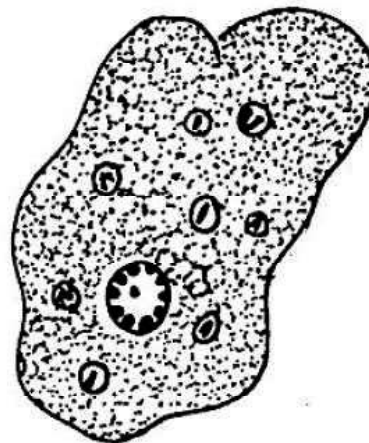
- By ingesting aquatic plants as food, which contains metacercarial kinds of the parasite that have come out of its intermediate host and encyst in aquatic plants.
- By skin transmission; for example, the filariform larvae of *Ancylostoma duodenale* and *Strongyloides stercoralis*, which abound in damp soil and may penetrate the skin when in contact with polluted soil.
- By contact with water containing the cercarial forms of *Schistosoma haematobium*, *S. mansonia* and *S. japoicum*, which may penetrate the skin of the person.
- By insects who host through blood-sucking arthropods that introduce the parasite directly into the blood or into the skin or into the skin layers at the time of obtaining a blood-meal; for example, *Plasmodium* the malarial parasite by the vector anopheline mosquitoes, *Trypanosoma* by glossina (Tsetse flies), *Leishmania* by phlebotomus (sandflies) and *Wuchereria* by culicine mosquitoes.
- Direct transmission by sexual contact as occurs with *monas vaginalis* or by kissing in the case of gingival amoebae.

Pathogenic Effects of Parasitic Infection in Man

The parasitic infections may remain unapparent or may cause disease of clinical significance. This depends on parasitic strain, size, number, metabolic process and host's age, immunity to infection, natural immunity, co-existing diseases, nutritional status and life style.

There are the following two kinds of parasitological infections:

- (i) **Protozoal infections:** These cause lesions by proliferation and multiplication and may get metastatically spread to distant organs. *E. histolytica* may act as a commensal multiplying in the intestinal lumen for long periods without invasion of tissue but its protozoal form secretes histolytic toxin, which lyses intestinal cells enabling the organism to penetrate the gut and spread to tissues.



Entamoeba

In plasmodium and other intracellular infections, the parasite, while multiplying within the cells (e.g., RBCs in plasmodium infection), destroys the cells.

- (ii) **Helminthic infections:** In such infections, human beings serve as definite hosts. Adult parasites are found inside the body and no multiplication takes place in humans except in *Strongyloides stercoralis* and *Hymenolepis nana* infections.

The clinical manifestations depend upon the number of invading organisms. They enter during primary infection and reinfection. The pathogenic and virulence of these helminths depends on their number, habitat and on the pattern of laying eggs or larvae and the products liberated from dead parasites in some cases.

Some specific examples are as follows:

- **Vascular or lymphatic obstruction:** Plasmodium falciparum infection can cause the blockage of capillaries of cerebral cortex that leads to fatal cerebral malaria. Elephantiasis by *Wucheria bancrofti* is a result of repeated lymphangitis, obstruction and lymphoedema.
- **Pressure effect:** Hydatid cyst causes pressure effects in surrounding tissue and the parasites, if and when present in vulnerable sites like the eyes and brain, may produce serious damage by pressure.
- **Physical obstruction:** If and when the masses of round worms occur, they can cause intestinal obstruction. One single worm may block the appendix or the bile duct.
- **Trauma:** Clinical disease resulting hookworm infections.
- **Allergic manifestations:** In some helminthic infections, the secretions and excretions of growing larvae and products released from dead parasites may give rise to allergic manifestations.
- **Predisposition to malignancy:** The parasitic infection may contribute to the development of neoplastic growth, e.g., liver flukes clonorchis and opisthorchis may induct the bile duct carcinoma and *Schistosoma haematobium* may contribute in bladder cancer.

Clinical Manifestations of Parasitic Infections

The following are the clinical manifestations of parasitic infections:

1. Elephantiasis caused by *Wucheria bancrofti* is an outcome of repeated lymph.
2. Plasmodium falciparum infection can cause the blockage of capillaries of cerebral cortex that leads to fatal cerebral malaria.
3. Hydatid cyst causes pressure in surrounding tissue; therefore, in vulnerable sites like eyes and brain, it may cause serious damage due to pressure.
4. If and when the masses of round worms occur, they can cause intestinal obstruction, which is also called physical obstruction.
5. Parasites may induce immune resistance in its host generating antibody response and T cell mediated immune response.

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Immunity in parasitic infection

Just as in case of other infectious agents, parasites can also induce specific immune resistance in its host. And these can generate antibody response and effector T cell (cell mediated) response. The immune elimination of parasite is much less efficient than that against bacterial and viral infections because the parasites have evolved to be closely adapted to the host, and majority of the parasitic infections are chronic and show a degree of host specificity.

Acquired immunity in parasitic infection

- Specific immune response to parasites results in antibody production of the immunoglobulin types IgG and IgM.
- IgE in helminthic infections and IgA is produced in intestinal protozoa (e.g., *Entamoeba* and *Giardia* spp.)
- The parasites are enormously large. Structurally and antigenically they are complex and therefore the specific antibodies provide several mechanisms of protection.
- Just like in viral infections, the anti-parasitic antibody is only effective against extracellular parasites and if parasitic antigens are exhibited by the infected cells on their surfaces.
- Neutralization occurs when the antibody combines with various surface molecules of parasites and then neutralizes them, blocking or interfering with their proper functioning. Antibody binds to toxins and enzymes of certain parasites. This protects the host from damage. It also affects the infection process.
- Agglutination of blood parasites by the IgM antibody may occur, which prevents the spread of the parasite as found in acute phase of infection with *Trypanosoma cruzi*.
- Antibodies can block the orifices of some worms. For example, oral and genital openings or it may interfere with the vital physiological functions thereby causing starvation or curtail reproduction.
- Opsonization: It is a situation in which an antibody acts as opsonin, thus increasing clearance by phagocytic cells and complement activation, resulting in the enhanced ingestion of organisms due to complement receptors.
- Antibodies may react with surface antigens, the surface of parasites, results in direct damage, or by interacting with complement-causing cell lysis.
- Antibody dependent cell-mediated cytotoxicity (ADCC): It is seen in *Trypanosoma cruzi*, *S. mansoni*, *Trichinella spiralis* and filarial worms. Effector cells, macrophages, monocytes, neutrophils and eosinophils combine with the iron and complement receptors of the antibody coated parasites and thus the effector cell and the target cell are required to be in close apposition because the toxic antigen antibody molecules produced are non-specific and could damage host cells.

Cellular response in parasitic infection

- T lymphocytes: CD4+ T cells act as helper cells in antibody production and CD8+ T cells are cytotoxic in several instances.
- Macrophages play a dominant role in the process of elimination of protozoa and worms.
- Granulocytes such as neutrophils and eosinophils also play important roles in the elimination of protozoa and helminths.

The complete elimination of infecting parasite followed by immunity to reinfection is rarely observed and immunity to reinfection in parasitic diseases lasts only so long as the original infection persists. After the complete elimination of a parasitic infection, either by natural means or therapy, the host remains susceptible to reinfection.

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3.2.2 Spread of Parasitic Infection and Host Parasite Relationship

Antonie van Leeuwenhoek was the first person who, with the help of a microscope having single lens, observed microscopic organisms in the faeces of man and animal and called them 'animalcules'. Between 1674 and 1716, he studied and gave a description of many free-living protozoa, which included the first parasitic protozoan named *Eimera stiedai*. He discovered the first human parasitic protozoan in 1681 and named it *Giardia lamblia*. He found the aforementioned protozoa in his own diarrhetic stools. Goldfuss, who belonged to Germany, was the first person who used the term protozoa. It was in 1782, when this parasite and its ova were first isolated. Thereafter, the introduction of Romanowsky stains in staining of blood smear brought a revolution in the study of parasites. During the 19th century, amoebae, trypanosomes and malaria parasites were documented.

Until the 1870s, the flukes or worms comprised almost all known parasites. In fact, the evidence of various worm infections is found in ancient Egyptian mummies (1210–1000 BC). In ancient Egypt, infections spread by worm were identified; these worms included *Taenia saginata*, *Ascaris lumbricoides*, *Schistosoma haematobium* (bilharzia) and *Dracunculus medinesis* (guinea worm). A current examination of the tissue sections of ancient Egyptian mummies revealed that *Trichinella spiralis* infection was prevalent during those days. Major historical discoveries of worms took place in 1379 (*Fasciola hepatica*) and in 1558 (*Cysticercus cellulosae*). During the latter part of the 19th century and early 20th century, their life cycle and related disease syndromes were discovered.

Today, parasitology is experiencing basic changes; exclusively, with the application of biochemical tools and molecular biology. These tools are increasingly being employed to diagnose the human parasites and evolve anti-parasitic vaccines in order to manage these infections.

The parasitic infections usually exist more in less developed areas, in warm climates and among the socially bereaved sections of a particular society. The economic and environmental factors are basically responsible for the endemicity of parasitic infections. Tools such as chemotherapy and vector control were identified as productive measures against the spread of malaria; however, they

had only short-term success as resistance against the drug has evolved among the mosquito vectors and the parasites. Modern immunological or chemotherapeutic methods may be required to contain these prominent diseases.

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Origin of Parasites

It is believed that their free-living ancestors gave birth to parasites, which also evolved along with their hosts. Identifying the evolutionary association between parasites and their hosts can offer important information about the interaction between the various groups of hosts.

The less developed monogenetic trematodes parasitize only fish. The highly developed digenetic trematodes are discovered in both fish and higher vertebrates; whereas, the much advanced digenetic trematodes parasitize the highest host groups.

Few parasites may need more than one group of hosts. In such cases, a definitive host harbours the adult stage of a parasite; whereas, an intermediate host harbours the larval forms. Few parasites may have multiple intermediate hosts.

A vector is a mollusc, arthropod or an agent, which transfers the parasite from one vertebrate host to another. If the parasite evolves within the vector, the vector is called biological vector; however, if parasite is passively transmitted by a vector, the vector is called a mechanical vector.

Host–Parasite Interaction

The entry of a particular parasite into a particular host is called parasitic infection, whose outcome decides the life of the parasite and development of disease.

Parasites may invade their host through following ways:

- By ingestion
- Through skin or mucous membranes
- Through the placenta from the mother during birth
- Through the transfusion of blood from donor to recipient

Once a parasite invades a host, it either gets killed or dies, or it may pass through the host unchanged. In any case, no prominent pathological conditions of infection are found in the host. The parasite may also live within the host and thus develop and multiply itself causing disease.

The difference between disease and an infection due to zooparasites must be understood here. There are various factors that determine the etiology or the cause of disease due to a parasite invasion of mammal hosts.

The host and parasite both contribute towards a very delicate balance, which either arrests infection or results in development of pathological conditions due to their particular nutritional or physiological needs. The mechanical destruction of host tissues can also cause disease. However, if the host tries to disallow the parasite attack through non-immunological and immunological ways, it leads to a chain of immunopathological reactions, which leads to illness. The intricacy of parasites' structure provides a major challenge to host immune responses; thus, the host–parasite interaction remains a dynamic process with the host using innate

and acquired immunological defence mechanisms. Maximum immunity is just extraordinarily defensive.

Parasites can enter the host's immune responses through the following ways:

- Changing their antigenic structure
- Intracellular location
- Change of the host response to favour the survival of the parasite

Table 1.1 gives details of the prominent parasitic protozoan and helminthic human infections, diseases, estimated prevalence and the major endemic areas. The amount of illness from these infections is cumbersome and the methods available for their control are very few. Table contains details of major arthropods, which are medically important as they act as vectors for parasitic or other infections.

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Table 3.1 Major Parasitic Infections of Man

| | Species | Disease | Estimated prevalence (millions) | Major endemic areas |
|--|--|--|---------------------------------|--|
| Protozoa | Plasmodium falciparum Plasmodium vivax Plasmodium malariae Plasmodium ovale | Malaria | 800 | Latin America, Africa, Asia |
| | Trypanosoma gambiense Entamoeba histolytica Tripanosoma rhodesiense Tripanosoma cruzi | African trypanosomiasis (sleeping sickness) Amoebiasis Chagas's disease Tripanosomiasis Chagas's disease | 20–40 Ubiquitous 40 | Tropical Africa Worldwide South America Central and South America |
| | Leishmania donovani | Kala-azar | 10–20 | Latin America Africa, Asia |
| | Leishmania tropica | Cutaneous leishmaniasis | | |
| | Leishmania mexicana Leishmania braziliensis | Mucocutaneous leishmaniasis | Ubiquitous | Worldwide |
| | Toxoplasma gondii | Toxoplasmosis | | |
| | Helminth | Ascaris lumbricoides Necator americanus Ancylostoma duodenale | Ascariasis Hookworm disease | 750 |
| Schistosoma haematobium | | Schistosomiasis | 200–300 | South America Caribbean, Africa, Asia |
| Schistosoma mansoni Schistosoma intercalasum Schistosoma mekongi | | (Bilharziasis) | | |
| Wucheria bancrofti Brugi malayi | | Lymphatic filariasis | 200–300 | |
| Onchocerca volvulus | | River blindness (onchocerciasis) | 50 | Central America West Africa Worldwide |
| Taenia solium Taenia saginata | | Teniasis | | |

Table 3.2 Major Arthropod Vectors of Human Parasitic Diseases

| Class | Genus | Etiological Agent |
|-----------|-------------|------------------------|
| Crustacea | Cyclops | Dracunculux medinensis |
| | Diaptomus | Diphyllbothrium latum |
| Insects | Anopheles | Plasmodia |
| | Triatoma | Trypanosoma cruzi |
| | Rhodnius | |
| | Phlebotomus | Leishmania |
| | Culex | Wucheria bancrofti |
| | Glossina | Trypanosoma |
| | Simulium | Onchocerca volvulus |

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Check Your Progress

1. Define parasites?
2. Which are the areas where parasitic infections are more prevalent?
3. Which parasites inhabit the surface of the body of the host without penetrating into the tissues?
4. What is meant by the indirect life cycle of a parasite?
5. Which are the factors that have a huge impact on parasitic infections in man?
6. What are the results of plasmodium falciparum infection?
7. Define a host.
8. What are the two kinds of parasitism?

3.3 PROTOZOAN PARASITES PATHOGENIC TO MAN

General Characteristics

The general characteristics of protozoa are as follows:

- They are eukaryotic microorganisms.
- They are unicellular and microscopic.
- They can move independently with the help of flagella, cilia and pseudopodia.
- They usually lack the capability for photosynthesis, although the *Euglena* genus is known for possessing the ability to conduct photosynthesis.
- They are known for reproducing by asexual methods, although sexual reproduction has also been observed in several species.
- Most protozoan species are aerobic, but some anaerobic species have also been found.
- They are found in most moist habitats, such as freshwater and marine environments. The terrestrial species are found in decaying organic matter and some are parasites of plants and animals.
- They vary substantially in size and shape, ranging from the size of fungal cells, visible only under a microscope to species visible to the naked eye.
- Most of their genera do not have cell walls and can take on a variety of shapes.
- Most of them have one nucleus, but some may have a macronucleus and one or more micronuclei.
- They obtain large food particles by phagocytosis, where the food is ingested into a vacuole and lysosomal enzymes digest the nutrients in the particle and the products are distributed in the cell.

Classification

Despite the major divisions, there is still a lot of confusion regarding the classification of protozoa. This is because of the lack of known specimens or the presence of strains or races of the same species with slightly different characteristics. The modes and mechanisms of reproduction have been used as the basis for determining the definitions of species. However, there are exceptions within these parasite groups. These organisms tend to alter their morphological forms according to age, host or nutrition. There are other problems also like assigning different names for the same organism for instance alternation of parasitic and free-living phases in the life cycle may lead to difficulties in recognizing organisms that belong to the same species.

The molecular methods of grouping organisms are important in confirming taxonomic conclusions and as studies continue in parasitic genetics, immunology and biochemistry, the species designation will become more defined. According to the Whittaker classification, protozoa belong to the Protista kingdom. They are categorized into different groups, mostly based on their motility. Some microbiologists refer to these groups as phyla and some refer to them as classes. The various members of the four major groups are discussed as follows:

- (i) **Mastigophora:** Mastigophora have one or more flagella (often 1–8; trypanosoma have one flagellum and giardia have eight flagella) and they reproduce usually by fission. Sexual reproduction is also observed in some of their species. The purpose of the flagellum is to either push the protozoan ahead or lead the cell along. These protozoa cause intestinal distress (*Giardia lamblia*) and sleeping sickness (*Trypanosoma brucei*) in humans. *Euglena gracilis*, also a part of this group, is a flagellated organism that possesses chlorophyll pigments used for photosynthesis.
- (ii) **Sarcodina:** This group consists of amoebas and their motility is assisted usually by pseudopodia. Asexual reproduction takes place by cellular fission. This group includes foraminiferans and radiolarians. The presence of these is marked by the presence of oil fields and marine geologists use them as potential markers for the same. *Entamoeba histolytica* is also a part of this group and the cause of amoebic dysentery.
- (iii) **Ciliophora:** This group consists of protozoa whose motility is assisted by cilia, whose rows vibrate in synchrony and push the organism in one direction. Both sexual reproduction and binary fission take place in this group. Paramecium and tetrahymena are members of this group.
- (iv) **Apicomplexa:** The members of this group form spores at one stage in their life cycle and are thus referred to as sporozoa. All species in this group are parasitic in nature and often cause malaria and toxoplasmosis. *Pneumocystis carinii* is an important pathogen of this group and causes lethal pneumonia in patients suffering from AIDS.

Life cycle of protozoa

A protozoa passes through several stages in its life cycle, and these stages differ in structure and activity.

Trophozoite is a generic term for the moving, feeding and reproducing stages of most protozoa. This stage is usually related with pathogenesis. In hemoflagellates,

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the terms amastigote, promastigote, epimastigote and trypomastigote depict the trophozoite stage, marked by the absence or presence of a flagellum. Other stages include the **merozoite** (resulting from asexual reproduction) stage and **gametocytes** and **gametes** (resulting from sexual reproduction) stages. Cysts are formed in some protozoa that contain one or more infective forms. These cysts multiply so that excystation discharges more than one protozoa.

Cyst maturity leads to nuclear division, the release of four uninucleate amoebas, doubling of organelles and forming of trophozoites. **Oocysts** is the name of the stage caused by sexual reproduction in *Apicomplexa*. Some apicomplexan oocysts are passed in the faeces of the host, but the oocysts of *Plasmodium* (malaria agent) develop inside the mosquito vector.

Growth and Nutrition Of Trophozoite Protozoa Nutrition

Trophozoites is the term used for the growing stage in the life cycle of protozoa, where they are actively feeding off the host and growing. They derive their nutrition by either diffusion or active transport across the plasma membrane. The larger-sized food particles are taken in the protozoa by the process of phagocytosis with the help of pseudopodia. Some species of protozoa possess a special mouth-shaped structure, also known as cytostomes, which help to ingest food. The smaller and minute droplets of food particles can enter the protozoan cells by pinocytosis. Protozoa respire by osmosis while excretion of waste products occurs with the help of contractile vacuoles.

Growth

Encystment: All protozoa can be transformed from an active or trophozoite stage to an inactive (or resting) or cystic stage that encloses itself within a tough wall. During the cystic stage, this parasitic protozoa loses its power of motility, growth and multiplication. In fact, the cyst stage is the resistant stage of the parasite that enables the organism to survive longer under unfavorable conditions.

Reproduction: In general, the protozoan parasite can exist in the trophozoite and cyst stages, which are both found in intestinal flagellates and amoebas. Here, the protozoa multiply only in the trophic form. Protozoa reproduction usually occurs asexually but sexual reproduction occurs in ciliates.

Sexual reproduction

Sexual reproduction may occur by either conjugation or syngamy.

- **Conjugation:** Two cells temporarily join together and reciprocally exchange nuclear material. Subsequently, the two cells separate. In fact, there is no increase in the number of cells. The process of conjugation is seen in ciliates like *Balantidium coli*.
- **Syngamy:** This is also known as gametogony where sexually differentiated male and female cells called gametes are produced. These can unite permanently resulting in complete fusion of their nuclear material, finally resulting in a resultant product that is called a zygote. This process is not seen in the flagellate plasmodium.

Asexual multiplication

It can occur by three methods that include the following:

1. **Binary fission:** This is the commonest method of multiplication and it occurs by mitotic division of the nucleus that is followed by division of the cytoplasm. During this process the individual protozoa divides either longitudinally as in flagellates, or transversely as in ciliates.
2. **Multiple fission:** This is also termed as schizogony where more than two individuals are produced as is seen in plasmodium. Here, the nucleus of the parent cell undergoes several successive divisions which are then surrounded by a portion of the cytoplasm that results in the formation of daughter cells termed as merozoites. After multiplication the parasitic body or schizont gets ruptured and it liberates daughter cells which once again turn repeat their life cycle.
3. **Endodyogeny:** In this method, the parasitic cell undergoes a single internal budding resulting in the formation of two daughter cells as seen in the genus *Toxoplasma*.

NOTES

Check Your Progress

9. Define parasitology.
10. Name two common protozoa parasites.
11. List the stages in the life cycle of protozoa.
12. How do protozoa respire?
13. What is Endodyogeny?

3.4 GEOGRAPHICAL DISTRIBUTION, HABITAT, MORPHOLOGY, LIFECYCLE, PATHOGENESIS, CLINICAL FEATURES, LAB DIAGNOSIS OF SOME PROTOZOAN PARASITES

You already know that protozoa are photosynthetic. Protozoa that use flagella as their mode of locomotion and reside in the intestine of the host are called intestinal flagellates.

In this section, we will discuss about the intestinal flagellates in detail, i.e. we will cover the morphology, pathogenesis, life cycle, clinical symptoms, diagnosis and treatment of the most common pathogenic intestinal flagellate – the *Giardia intestinalis* or *Giardia lamblia*. Other intestinal flagellates include *Chilomastix mesnili* (non-pathogenic), *Trichomonas* spp. and *Dientamoeba fragilis*. Table 3.3 lists various types of protozoan diseases, their causal agents and the organs affected.

Table 3.3 Important Infections Caused by Protozoa

NOTES

| Name of Disease | Causal Agent | Organs Affected |
|-------------------------------------|------------------------------|-------------------------------------|
| Amoebiasis | <i>Entamoeba histolytica</i> | Intestine, liver |
| Primary amoebic meningoencephalitis | <i>Naegleria fowleri</i> | Brain, lungs |
| Giardiasis | <i>Giardia lamblia</i> | Intestines |
| Trichomoniasis | <i>Trichomonas vaginalis</i> | Urogenital organs |
| African sleeping sickness | <i>Trypanosoma brucei</i> | Blood, brain |
| Leishmaniasis | <i>Leishmania donovani</i> | White blood cells, skin, intestines |
| Toxoplasmosis | <i>Toxoplasma gondii</i> | Blood, eyes |
| Malaria | <i>Plasmodium species</i> | Liver, red blood cells |
| Babesiosis | <i>Babesia microti</i> | Red blood cells |
| Pneumocytosis | <i>Pneumocystis carinii</i> | Lungs |

Some of the diseases caused by Protozoa are discussed below:

1. Giardiasis

Morphology

This is an intestinal flagellate belonging to the order Diplomonadida. This protozoan was named *Cercomonas intestinalis* by Lambl in 1859. *Giardia lamblia* was the name given by Stiles in 1915, in the honour of Professor A. Giard of Paris and Dr. F. Lambl of Prague.

- The protozoan is pear-shaped, with a broad anterior and narrow posterior.
- It has a bilateral symmetry and is 10–15 μm long. Just like a suction pump, the protozoan uses its large adhesive disk, present on the anteroventral side, to attach to the mucosa of the host.
- There are four pairs of flagella: one anterior pair, two posterior pairs and a caudal pair.
- It bears two nuclei containing a dark endosome and has no mitochondrion, endoplasmic reticulum, golgi or lysosomes.

Figure 3.1 will give you a clear idea of what *Giardia* looks like.

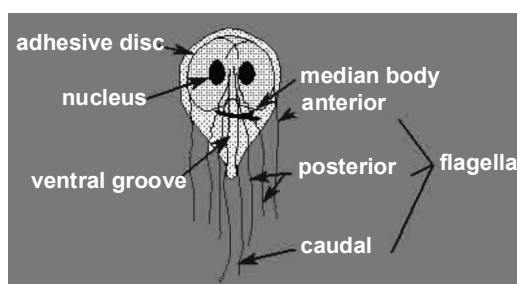


Fig. 3.1 Structure of Giardia

(Source: <http://microbewiki.kenyon.edu/index.php/Giardia>)

Pathogenesis and Life Cycle

Infection with *Giardia lamblia* may result into asymptomatic to severe giardiasis.

Now, let us read about the life cycle of this pathogen.

- You will find two stages in the life cycle of *Giardia*: trophozoite and cyst.
- Cysts are the resistant forms. They are responsible for the transmission of giardiasis. They can survive several months in cold water. Infection is by the ingestion of cysts in contaminated water, food or faecal–oral route.
- The cysts, after reaching the small intestine release trophozoites (10–20 μm x 5–15 μm) in the duodenum with the help of multiple binary fission.
- The trophozoites attach to the intestinal epithelium using their adhesive disk and then reproduce by binary fission (refer Figure 3.2).

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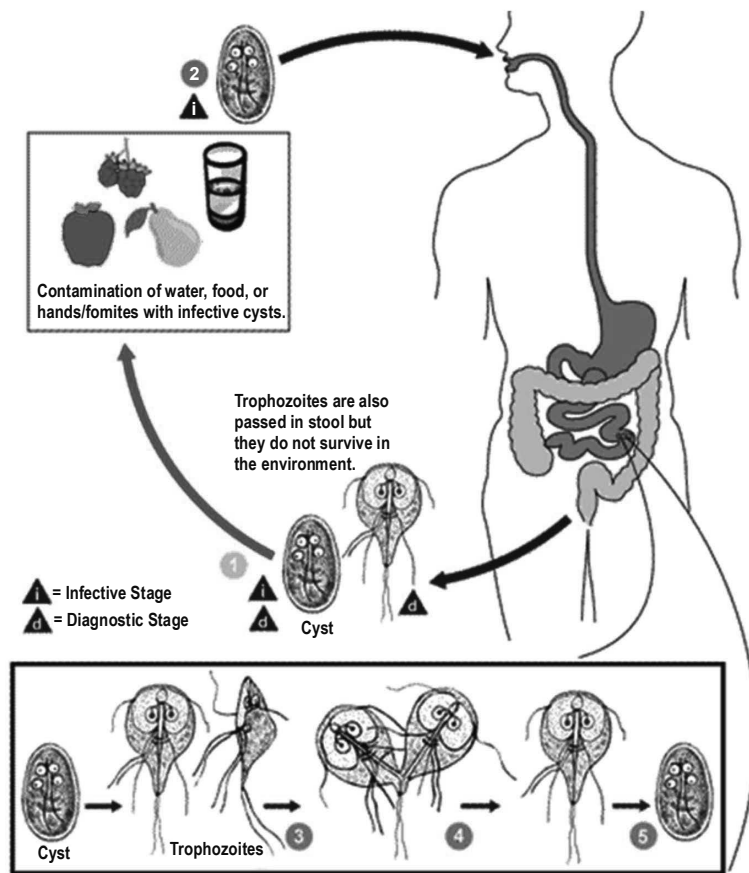


Fig. 3.2 Pathogenesis of *Giardia intestinalis*

(Source: <http://www.dpd.cdc.gov/dpdx>)

Clinical Manifestation

- Cysts are infectious when passed in the stool.
- Acute giardiasis develops after an incubation period of 1–14 days and usually lasts for 1–3 weeks.
- You may have seen people with symptoms like diarrhoea, abdominal pain, bloating, nausea and vomiting. These symptoms may indicate the acute form of giardiasis.
- In chronic giardiasis, the symptoms are recurrent and malabsorption and debilitation (serious weakening and loss of energy) may occur. Pus, mucous and blood are occasionally present in stools.

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Diagnosis

- Giardiasis is diagnosed by the identification of cysts or trophozoites in the faeces, using direct mounts as well as concentration procedures.
- Repeated samplings may be necessary. In addition, samples of duodenal fluid (e.g., Enterotest) or duodenal biopsy may demonstrate trophozoites.
- Alternate methods for detection include antigen-detection tests by enzyme immunoassays and detection of parasites by immunofluorescence. Both methods are available in commercial kits.

Treatment: For the treatment of giardiasis, the doctor will prescribe **metronidazole** (dosage: 250 mg per oral three times a day, for 5–7 days for adults and 5 mg/kg per oral three times a day, for 5–7 days for children) and **tinidazole** (dosage: 2 g per oral once with food for adults and 50 mg/kg per oral once with food for children older than 3 years).

Trypanosoma, Leishmania and Toxoplasma: Morphology, Pathogenesis, Clinical Manifestation and Diagnosis

In the previous section, we discussed about the most common intestinal protozoa, i.e. *G. lamblia*. You will now learn that some protozoa may also live and grow inside the blood. *Trypanosoma* and *Leishmania* are such haemoflagellates and belong to the Trypanosomatidae family. Several species of these genera are important pathogens. They are parasitic and are found in the blood of many species of domestic and wild animals, birds and humans. *Toxoplasma* belongs to the Sarcocystidae family.

2. *Trypanosoma*

Trypanosoma is a genus of kinetoplastids (class Kinetoplastida), a monophyletic group of unicellular parasitic flagellate protozoa. All trypanosomes are heteroxenous and are transmitted via a vector. The different trypanosomes include the following:

- (i) *Trypanosoma brucei*: *T. b. brucei*, *T. b. gambiense* and *T. b. rhodesiense*
- (ii) *Trypanosoma cruzi*

A. Morphology

To identify the parasite, it is very important that we understand what it looks like.

- A trypanosome is an elongated cell that has a single nucleus that usually lies near the centre of the cell that bears a single flagellum.
- The flagellum, whose length and position on trypanosome is variable, begins from a kinetoplast, which is a specialized part of the mitochondria (refer Figure 3.3).
- When recovered from a blood-infected person, this flagellum originates near the posterior end of the cell and then passes over the cell surface.
- The sheath forms a wavy flange, namely, undulating membrane.
- Morphologically, you will come across three types of trypanosomes in blood, which are as follows:
 - o **The slender forms:** These are long and thin, roughly 29- μ m long with a free flagellum.

- o The **stumpy forms**: These are thick and short, approximately 18- μm long with no free flagellum.
- o The **intermediate forms**: These are 23- μm long with a moderately thick body and a free flagellum of medium length.

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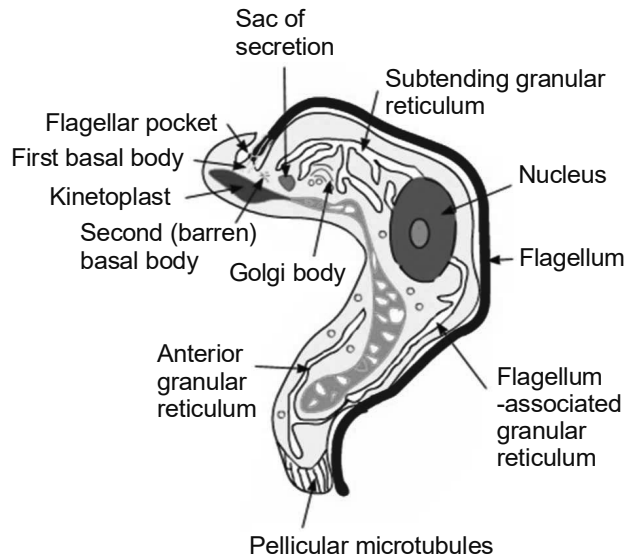


Fig. 3.3 Morphology of Trypanosoma

(Source: <http://pathmicro.med.sc.edu/lecture/diag-try4.jpg>)

B. Pathogenesis

Trypanosomes are meta-cyclic and are transmitted by inoculation through proboscis of the insect vector. The disease is a zoonotic one that spreads from animal to man. *Trypanosoma brucei brucei* and *T. b. rhodesiense* cause acute sleeping sickness, while *T. b. gambiense* causes chronic sleeping sickness. *T. cruzi* is a metacyclic trypanosome found in the posterior gut of the insect vector. It causes American trypanosomiasis (Chagas disease).

To understand the pathogenesis well enough, it is very important that we understand the life cycle of this parasite:

- Meta-cyclic trypomastigotes, found in the faeces of a reduviid bug that has fed on the human skin, gets rubbed into the human skin, either through scratching or penetration of the conjunctiva or membranes of the nose and the mouth.
- The trypomastigotes transform to become amastigotes in the reticuloendothelial cells and start multiplying.
- The amastigotes get released into the blood.
- The trypomastigotes reach cells of heart muscle, nerves, skeletal muscle and smooth muscle of the gastrointestinal tract through blood and lymphatic system.
- The amastigotes multiply, forming pseudocysts.
- Some amastigotes in the pseudocysts get elongated and develop into epimastigotes.

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- Epimastigotes get transformed to trypomastigotes.
- Upon cell rupture, the trypomastigotes get released into the blood.
- The life cycle is complete with a triatomine bug vector ingesting circulating trypomastigotes. The trypomastigotes transform into epimastigotes. They start multiplying in the gut of the bug by binary fission.

Now, let us see how *T. cruzi* invades its host and causes American trypanosomiasis. See Figure 3.4 for a better understanding of the life cycle of *T. cruzi*.

Triatomine bugs (*Panstrongylus megistus*, *Triatoma infestans* and *Rhodnius prolixus*) are the intermediate hosts of *T. cruzi*. Humans, however, are the definitive hosts of *T. cruzi*. The life cycle of *T. cruzi* consists of the following stages:

- Humans get infected when the reduviid bug bites them. With the bite, the bug discharges its infected faeces near the bite wound, which then get rubbed onto the wound.
- Meta-cyclic trypomastigotes invade the reticuloendothelial system and spread to tissues, where they get converted into amastigote forms.
- Amastigotes multiply by binary fission and develop into trypomastigote forms.
- Trypomastigotes get liberated into blood. Now, when a reduviid bug bites a person, the trypomastigotes are taken up with its blood.
- These trypomastigotes transform to amastigotes in the foregut and then in the midgut get converted into epimastigotes. These develop into metacyclic trypomastigotes in the hindgut of the bug and are excreted in the faeces of the bug.
- These infected bugs bite new definitive host, and the cycle gets repeated.

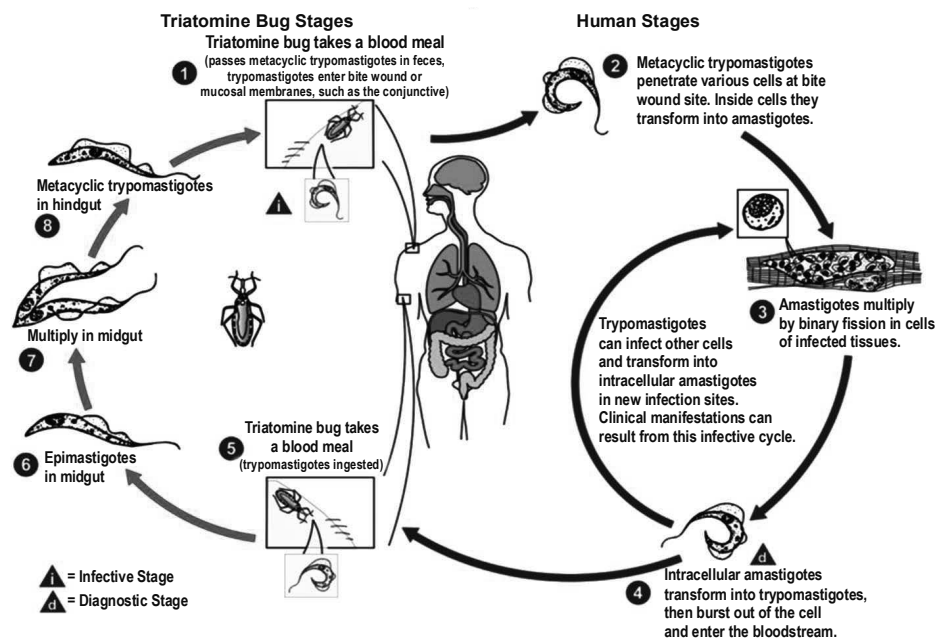


Fig. 3.4 Life Cycle of *T. cruzi*

C. Clinical manifestation

Since different species of *Trypanosoma* are responsible for causing different diseases, let us discuss the symptoms of these diseases one by one.

- (i) **Sleeping sickness (African trypanosomiasis):** The general symptoms include anxiety, drowsiness during the day, fever, headache, insomnia at night, mood changes, sleepiness (may be uncontrollable), sweating, swollen lymph nodes all over the body, swollen, red, painful nodule at site of fly bite and weakness.
- (ii) **Chagas disease (American trypanosomiasis):** During the acute phase of the disease, symptoms are generally absent or mild. Fever, headache, lymphadenopathy, pallor, muscle pain, difficulty in breathing, swelling and abdominal or chest pain may occur. During the chronic phase, patients may experience cardiac disorders and up to 10 per cent patients suffer from digestive, neurological or mixed ailments. Just like any other disease worsens if not cured in time, this disease may also prove to be fatal. At later stages, the infection can lead to sudden death or heart failure caused by progressive destruction of the heart muscle.

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D. Laboratory diagnosis

- (i) **Specimen:** For the diagnosis of trypanosomiasis, you may take blood, cerebrospinal fluid (CSF) aspirate or tissue material as a specimen.
- (ii) **Microscopic examination:** For microscopic examination of the parasite, stain the blood film with Giemsa stain. You will see typical trypomastigotes in a C-shaped form.
- (iii) **Culture:** You can culture *T. cruzi* on NNN (Novy, McNeal and Nicolle) medium when incubated at 22–24°C.
- (iv) **Serology:** If you want to diagnose chronic cases, where detection of the parasite is quite difficult, serology can prove to be a useful technique. In this, *T. cruzi* antibodies are detected in the serum of the patient by the Machado–Guerreiro test, indirect fluorescent antibody test; indirect haemagglutination assay and enzyme-linked immunosorbent assay (ELISA).
- (v) **Antigen detection:** Antigens can be detected in the urine and sera of patients with chronic Chagas disease.
- (vi) **Skin test:** The extracts of *T. cruzi* culture are used for intradermal inoculation, and a delayed type of hypersensitivity reaction is noted in positive cases.
- (vii) **Molecular tests:** Polymerase chain reaction (PCR) may be employed for detecting chronic Chagas disease in patients with very few trypomastigotes in the blood.
- (viii) **Histopathological examination:** Biopsy from lymph node or tissue reveals amastigote forms of *T. cruzi*.

3. *Leishmania*

Leishmania is a genus of trypanosomatid protozoa. It is the parasite that is responsible for the disease leishmaniasis. It commonly infects hyraxes, canids,

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rodents and humans. If a sand fly (genus *Phlebotomus*, subfamily Phlebotominae) carrying this parasite bites you, you may get infected with this disease. About 20 species of *Leishmania* cause human infection. These include the *L. donovani* complex with three species (*L. donovani*, *L. infantum* and *L. chagasi*), the *L. mexicana* complex with three main species (*L. mexicana*, *L. amazonensis* and *L. venezuelensis*), *L. tropica*, *L. major*, *L. aethiopica* and the subgenus *Viannia* with four main species [*L. (V.) braziliensis*, *L. (V.) guyanensis*, *L. (V.) panamensis* and *L. (V.) peruviana*].

A. Morphology

- *Leishmania*, in general, are seen as flagellated extracellular promastigotes in the sand fly vector (refer Figure 3.5).
- You can see them as aflagellar (without flagella) and as obligate intracellular amastigote forms within the mononuclear phagocytes of their vertebrate hosts.
- You cannot distinguish different species morphologically from one another.
- When they are stained with Romanowsky stains like as Giemsa, they appear as round or oval bodies with a well-defined nucleus and kinetoplast.
- The kinetoplast is a rod-shaped specialized mitochondrial structure that contains extranuclear DNA.

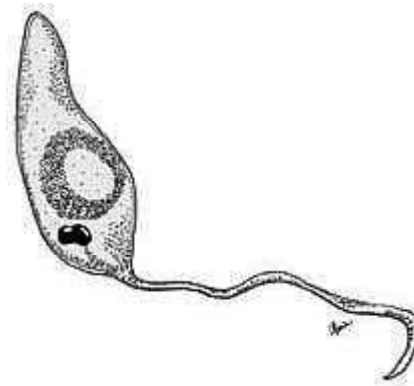


Fig. 3.5 Promastigote form of *Leishmania*

(Source: <http://www.cdc.gov/healthypets/diseases/leishmania.htm>)

B. Pathogenesis and clinical manifestation

Different species of *Leishmania* cause different types of leishmaniasis. Let us discuss the pathogenesis and symptoms of these types one by one.

- (i) **Visceral leishmaniasis:** It is the most severe form and may prove fatal if untreated. It is also known as kala-azar and is caused by the *L. donovani* complex.

Clinical description of visceral leishmaniasis (refer Figure 3.6): The infection is usually insidious with fever, sweating, weakness and weight loss with fever, hepatosplenomegaly and anaemia. It affects liver, spleen and bone marrow. Liver enlarges due to hyperplasia of Kupffer cells, which are packed with amastigotes. There is infiltration of bone marrow with parasitized

macrophages. Death occurs as a result of secondary bacterial infection, such as pneumonia or tuberculosis.



Fig. 3.6 A Boy Suffering from Kala-azar

(Source: <http://www.stanford.edu/class/humbio103/ParaSites2006/Leishmaniasis/visceral.htm>)

- (ii) **Cutaneous leishmaniasis:** It is the most common form, causing a sore at the bite site. This form may lead to any of the other three forms. It is caused by *L. tropica*, *L. major*, *L. aethiopica* and *L. mexicana*. The cutaneous leishmaniasis may also be of diffuse type, in which there are widespread skin lesions resembling leprosy. It is difficult to treat them.

Clinical description of skin or cutaneous leishmaniasis: When an infected sand fly bites, there appears a small red papule at the site of the bite. The patient mounts hypersensitive response. The papule eventually ulcerates and becomes depressed. It then heals, leaving a scar. The patient becomes immune to bites.

- (iii) **Mucocutaneous leishmaniasis:** It is caused by *L. braziliensis*. It occurs with skin ulcers, which spread, causing damage to the tissues of nose and mouth (refer Figure 3.7).

Clinical description of mucocutaneous leishmaniasis: It is called espundia. In the beginning, it develops as cutaneous leishmaniasis but eventually develops into lesions in the mucocutaneous junction of the pharynx. Sometimes, it also affects the genitalia or anus. The lesions may lead to deformities of the nose and mouth.



Fig. 3.7 A Boy Suffering from Mucocutaneous Leishmaniasis

(Source: <http://www.stanford.edu/class/humbio103/ParaSites2006/Leishmaniasis/Mucocutaneous.htm>)

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Life cycle of *Leishmania*

We now know about the symptoms of various forms of leishmaniasis. But, have you ever wondered how such serious infections occur just by a bite of a sand fly? Let us see what the life cycle of this parasite looks like.

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Stage 1: Leishmaniasis is transmitted by the bite of infected female phlebotomine (relating to sand flies of the genus *Phlebotomus*) sand flies, which inject the infective promastigotes from their proboscis during blood meals.

Stage 2: The promastigotes that reach the puncture wound are phagocytized by macrophages and other mononuclear phagocyte cells.

Stage 3: Promastigotes get transformed in these cells into amastigotes, which are the tissue-stage parasites.

Stage 4: They multiply by simple division and then move to infect other mononuclear phagocytic cells.

Stage 5: Host–parasite interactions at this stage decide whether this infection becomes symptomatic and cutaneous or visceral leishmaniasis.

Stage 6: New sand flies become infected by ingesting these infected cells during their blood meals.

Stage 7: Here the amastigotes transform into promastigotes.

Stage 8: These promastigotes develop in various regions of the gut (hindgut for leishmanial organisms in the *Viannia* subgenus; in the midgut for organisms in the *Leishmania* subgenus) and then migrate into the proboscis of the sand fly to complete the cycle. For your better understanding, we have illustrated the life cycle in Figure 3.8.

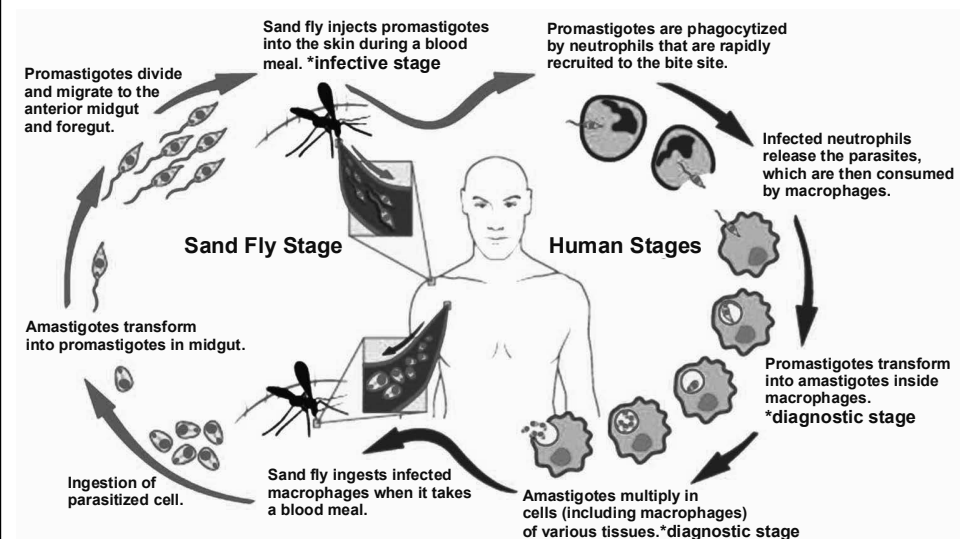


Fig. 3.8 Life Cycle of *Leishmania*

D. Laboratory diagnosis of *Leishmania* infections

- You can isolate the parasite of visceral leishmaniasis or kala-azar from suspected material, such as blood or biopsy content.
- *Leishmania* antibodies are demonstrated in the serum of patient.
- Non-specific tests—based upon the alterations in the normal blood picture and the serum proteins—can be conducted. These tests include progressive neutropaenia with relative lymphocytosis and monocytosis, in which leukocyte count progressively decreases as the disease progresses, and a WBC ratio ranges from 1:1000 to 1:2000.
- Sometimes, it becomes very difficult to diagnose the actual disease and there are cases when the patient keeps receiving treatments for various other diseases. Same can be the case with kala-azar patients, as this disease resembles many other diseases. A doctor may give various treatments to the patient for diseases like malaria, typhoid fever, amoebiasis, pulmonary tuberculosis, brucellosis, etc. before actually diagnosing kala-azar.
- There is a reversal of albumin:globulin ratio due to increase of globulin amounts in serum. This is detected by the following tests:
 - (i) **Napier's aldehyde test:** You can perform this test by taking one millilitre of patient's serum and mixing it with a drop of 40 per cent formalin. Shake the mixture and keep it at room temperature. Also, set up a control tube with normal serum. Gellification indicates a positive reaction, seen by opacification of the patient's serum within 2–20 minutes of the test. If you are performing this test on patients with disease of 3- to 4-month duration, this test will come out to be positive in 80–85 per cent cases. But, it is non-specific as other conditions like multiple myeloma, cirrhosis of liver and schistosomiasis also depict similar findings.
 - (ii) **Chopra's antimony test:** Take 1–2 ml of patient's serum in a test tube. Add a few drops of 4 per cent urea stibamine solution to it. A flocculent precipitate will indicate a positive test.
 - (iii) **Serum electrophoresis:** It determines whether a patient is suffering from hypergammaglobulinaemia. Hypergammaglobulinaemia refers to a condition that is characterized by the increased levels of a certain immunoglobulin in the blood serum of the patient. The name of the disorder refers to the position of the excess of proteins after serum protein electrophoresis (found in the gammaglobulin region).
 - (iv) **L–D bodies:** You can obtain the clinical specimens, such as splenic aspirates, bone marrow, etc. from a person with visceral leishmaniasis. (Splenic aspirate is the optimum specimen if the patient maintains an adequate prothrombin level.) Ensure that the platelet count of the patient is enough for aspiration. Inoculate the specimen into the water of condensation in protozoan culture media. Incubate this culture at 24°C for seven days. The parasites will grow as promastigotes, which are demonstrated in stained or unstained smears. The sample is recorded as negative after 5–6 weeks of incubation.

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Animal inoculation: The material from patients is injected intraperitoneally into hamsters or mice. These animals are kept at 23–26°C. Clinical specimens are taken at autopsy, demonstrating amastigotes or L–D bodies.

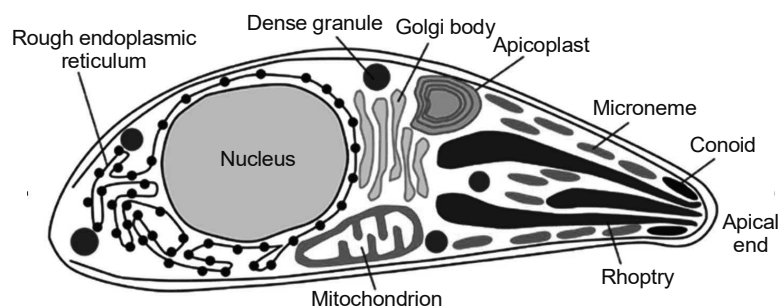
- (v) **Leishmanin or Montenegro test:** It is a skin test based on type 4 hypersensitivity or delayed hypersensitivity. An **antigen suspension** of washed promastigotes (0.1 ml quantity) prepared in 0.5 per cent phenol saline is injected intradermally on the dorsoventral region of forearm of a suspected person. A positive finding is indicated by induration of 5 mm or more in 48–72 hours at the site of injection. This test is used for epidemiological susceptibility to leishmaniasis, but it is negative in kala-azar.

4. *Toxoplasma*

Toxoplasmosis is considered to be the leading cause of death attributed to food-borne illness, but very few have symptoms because the immune system usually keeps the parasite from causing illness. Toxoplasmosis is considered one of the neglected parasitic infections. Women newly infected with *Toxoplasma* during pregnancy and anyone with a compromised immune system should be aware that toxoplasmosis can have severe consequences.

A. Morphology

- You will find several distinct stages in the life of *T. gondii*: rapidly dividing tachyzoites (in pseudocysts) (refer Figure), slow growing bradyzoites (in tissue cysts), and sporozoites (in oocysts).
- Individual organisms are crescent shaped, about $6 \times 2 \mu\text{m}$, and multiply within host cells. *Toxoplasma* reproduces sexually only in cats.
- Organisms infecting the intestinal epithelium produce oocysts, which are shed in the faeces.
- Mature oocysts are $\sim 12 \mu\text{m}$ in diameter and contain eight infective sporozoites.



Ultrastructure of *T. gondii* Tachyzoite

(Source: http://en.citizendium.org/wiki/toxoplasma_gondii)

B. Pathogenesis and risk factors

Toxoplasmosis is caused by the protozoan parasite *Toxoplasma gondii*. In various places throughout the world, it has been shown that up to 95 per cent of some

populations have been infected with *Toxoplasma*. Infection is often highest in those areas of the world that have hot, humid climates and lower altitudes. Toxoplasmosis is not passed from person to person, except in instances of mother-to-child (congenital) transmission and blood transfusion or organ transplantation. People typically become infected by three principal routes of transmission.

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(i) Food-borne transmission

- Eating undercooked, contaminated meat (especially pork, lamb and venison)
- Accidental ingestion of undercooked, contaminated meat after handling it and not washing hands thoroughly (*Toxoplasma* cannot be absorbed through intact skin)
- Eating food that was contaminated by knives, utensils, cutting boards or other foods that had contact with raw, contaminated meat

(ii) Animal-to-human (zoonotic) transmission

- Cats become infected by eating infected rodents, birds or other small animals.
- The parasite is then passed in the cat's faeces in an oocyst form, which is microscopic.
- They can shed millions of oocysts in their faeces for as long as 3 weeks after infection.
- Mature cats are less likely to shed *Toxoplasma* if they have been previously infected.
- A *Toxoplasma*-infected cat that is shedding the parasite in its faeces contaminates the litter box. If the cat is allowed outside, it can contaminate the soil or water in the environment as well.

(iii) Mother-to-child (congenital) transmission

- A woman who is newly infected with *Toxoplasma* during pregnancy can pass the infection to her unborn child (congenital infection).

(iv) Rare instances

- Organ transplant recipients can become infected by receiving an organ from a *Toxoplasma*-positive donor.

C. Aetiology and life cycle

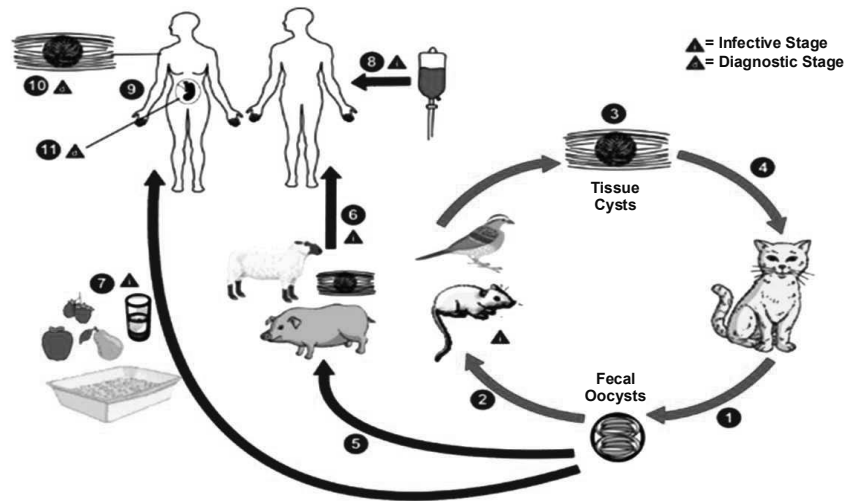
T. gondii is a protozoan parasite that infects most species of warm-blooded animals, including humans, and can cause the disease toxoplasmosis.

The only known definitive hosts for *T. gondii* are members of the family Felidae (domestic cats and their relatives). The life cycle of *T. gondii* begins when unsporulated oocysts are shed in cat's faeces. You will see in Figure 3.10 how the life cycle of this pathogen proceeds. This has also been detailed in points below.

1. Although oocysts are usually only shed for 1–2 weeks, large numbers may be shed. Oocysts take 1–5 days to sporulate in the environment and become infective. Intermediate hosts in nature (including birds and rodents) become infected after ingesting soil, water or plant material contaminated with oocysts.

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2. Oocysts transform into tachyzoites shortly after ingestion. These tachyzoites localize in neural and muscle tissue and develop into tissue cyst bradyzoites.
3. Cats become infected after consuming intermediate hosts harbouring tissue cysts.
4. Humans can become infected by several routes. In the human host, the parasites form tissue cysts, most commonly in skeletal muscle, myocardium, brain and eyes; these cysts may remain throughout the life of the host.



Life Cycle of *T. gondii*

(Source: <http://www.dpd.cdc.gov/dpdx>)

D. Clinical manifestation

- The parasite is responsible for tissue damage in its tachyzoite stage.
- The clinical symptoms and seriousness of the disease depend on the number of tachyzoites present, the ability of the host immune system to fight these tachyzoites, and the organs damaged by them.
- In young animals, like puppies, kittens, and piglets, tachyzoites spread via bloodstream and cause interstitial pneumonia, myocarditis, hepatic necrosis, meningoencephalomyelitis, chorioretinitis, lymphadenopathy and myositis.
- Other symptoms may range from fever, diarrhoea, cough, dyspnoea, icterus and seizures to death. Abortion and stillbirth in sheep and goats may be a result of *T. gondii* infection. Tachyzoites may also infect the foetus, causing necrosis in multiple organs.
- Finally, immunocompromised adult animals (e.g., cats infected with feline immunodeficiency virus) are extremely susceptible to developing acute generalized toxoplasmosis.
- In humans, the infection is often asymptomatic. Fever, lymphadenopathy, muscle aches may be present in immunocompetent individuals and encephalitis may be present in immunosuppressed patients.
- Congenitally infected children may suffer from impaired vision and mental retardation.

- The infection is also known to be associated with behavioural changes, including slower reaction times, increased risks of traffic accidents and suicidal behaviours.
- The infection has also been connected with schizophrenia, hallucinations and reckless behaviour.

E. Laboratory diagnosis

- The diagnosis of toxoplasmosis is typically made by serologic testing. A test that measures immunoglobulin G (IgG) is used to determine if a person has been infected. If it is necessary to try to estimate the time of infection, which is of particular importance for pregnant women, a test which measures immunoglobulin M (IgM) is also used along with other tests such as an avidity test.
- You can also diagnose the parasite by direct observation in stained tissue sections, cerebrospinal fluid (CSF) or other biopsy material. These techniques are used less frequently because of the difficulty of obtaining these specimens.
- Parasites can also be isolated from blood or other body fluids (for example, CSF), but this process can be difficult and requires considerable time.
- You may find molecular techniques, which detect the parasite's DNA in the amniotic fluid, useful in cases of possible mother-to-child (congenital) transmission.
- Ocular disease is diagnosed based on the appearance of the lesions in the eye, symptoms, course of disease, and often serologic testing.

Pathogenic Free-Living Amoebae

In all the previous sections, we learnt about various flagellated protozoa. However, this section deals with protozoa without flagellum. These protozoa move with the help of pseudopodium and are called amoebae. There are many free-living amoebae, but only few are known to cause infections in humans, e.g., *Naegleria fowleri*, *Acanthamoeba* spp., *Ballamuthia mandrillaris* and *Sappinia pedata*. These amoebae are ubiquitous and are found in soil, water, dust, air, etc. They live as parasites when they invade humans.

5. Morphology of *Naegleria fowleri*

- You will find three stages in the life cycle of *N. fowleri*: trophozite (amoeba), flagellate, and cyst stage. In all three stages, there is one nucleus with a large karyosome and no peripheral chromatin.
- Trophozites are about 10-20 μm in diameter, have a slug-like, shape and are mobile. Movement occurs with the help of blunt pseudopodia called lobopodium. Trophozites have mitochondria and demonstrate aerobic metabolism. The flagellate is pear shaped and has two flagella.
- Presence of the flagellar stage is a distinguishable feature of *N. fowleri* when compared with other pathogenic free-living amoebae, which do not have a flagellate form.
- The spherical cyst is single-walled and 8–12 μm in diameter. The amoeba encysts under harsh conditions.

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- Figure 3.11 shows you the structure of *N. fowleri* trophozoite.

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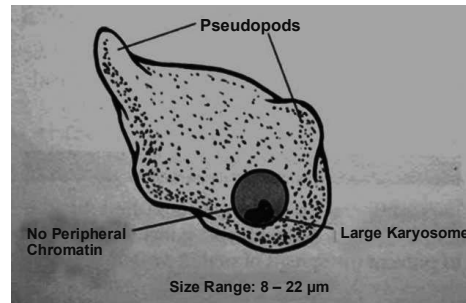


Fig. 3.11 Trophozoite of *N. fowleri*

(**Source:** <http://www.studydroid.com/printerFriendlyViewPack.php?packId=108146>)

Pathogenesis

- Primary amoebic meningoencephalitis (PAM) occurs in Europe and North America from amoebic invasion of the brain. The free-living soil amoebae *Naegleria fowleri*, *Acanthamoeba castellanii* and, possibly, species of *Hartmannella* have been implicated. Most cases have developed in children who were swimming and diving in warm, soil-contaminated pools, either indoors or—usually—outdoors.
- The amoebae, primarily *N. fowleri*, apparently enter via the nose and the cribriform plate of the ethmoid, passing directly into brain tissue, where they rapidly form nests of amoebas that cause extensive haemorrhage and damage, chiefly in the basilar portions of the cerebrum and the cerebellum. In most cases, death ensues in less than a week.
- Entry of *Acanthamoeba* into the central nervous system from skin ulcers or traumatic penetration, such as keratitis from puncture of the corneal surface or ulceration from contaminated saline used with contact lenses, has also been reported.

Clinical Manifestation

PAM is accompanied by high fever, headache, photophobia, stiff neck, nausea and vomiting. Other symptoms are confusion, drowsiness, seizures and coma. The infection may progress rapidly. Rarely, patients with PAM may experience disordered smell or taste.

Diagnosis

- **Specimen:** Cerebrospinal fluid
- **Microscopic examination:** If you want to diagnose this disease, you will need to do the microscopic examination of the cerebrospinal fluid. You will observe trophozoites and red cells but no bacteria.
- **Culture:** Amoebae can be readily cultured on non-nutrient agar plates seeded with *Escherichia coli*. These soil amoebas are distinguished by a large, distinct nucleus; by the presence of contractile vacuoles and mitochondria (absent in entamoeba) and by the presence of cysts that have a single nucleus

and lack glycogen or chromatoidal bodies. *Acanthamoeba* may encyst in invaded tissues, whereas *Naegleria* does not.

If a person suffering from PAM goes to a doctor, he would probably recommend amphotericin B. It has been successful in a few cases, chiefly in the rare instances when diagnosis can be made quickly.

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3.4.1 Protozoan Parasites belong to Sporozoa and Ciliata

1. *Plasmodium*

Plasmodium is a common spore-forming sporozoan parasite on human which causes malaria. Until 1935, malaria was a common disease in humans creating a serious problem and causing death *en masse*. But with the development of awareness and research programmes, antimalarial drugs were formulated. Still in African and Asian countries including India, malaria has not been fully eradicated despite of efforts from Government organizations. As per recent estimates about 300 million people suffer from malaria and about 2-4 million die each year.

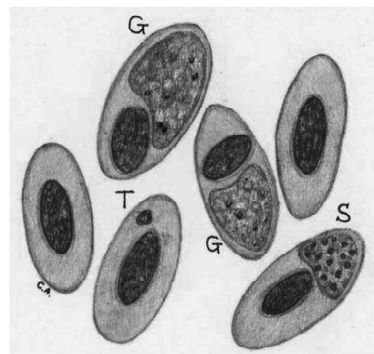


Fig. 3.12

Four pathogenic species of *Plasmodium* viz., *P. vivax*, *P. falciparum*, *P. malariae* and *P. ovale* have been recognised. *P. falciparum* is the most dangerous species followed by *P. vivax*.

A. Life Cycle

Malaria is associated with a mosquito vector, *Anopheles*. The life cycle of *Plasmodium* is very complex. The mosquito carries the inocula of *P. vivax* where it multiplies and produces the sporozoites. After mosquito bite, the sporozoites present in saliva are delivered in the blood of human. Thirty minutes after bite, the sporozoites enter in liver cells and undergo reproductive schizogamy through a series of intermediate stages. At the last stage of schizogamy, a large number of merozoites are released in the blood stream. The merozoites are infectious which in turn infect the red blood corpuscles and undergo reproductive schizogamy. A large number of merozoites are released upon rupture of RBCs. The merozoites infect the other RBCs and renew the cycle.

The infected RBCs are disintegrated which release the toxic substances. The toxic substances induce the characteristic symptoms of malaria i.e. chills and fever. After reaching the body temperature upto 40°C, fever subsides. However, due to break down of RBCs, the patient suffers from anaemia, and liver and

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spleen enlargement. As per estimate 1% infected RBCs contain about 10,000 millions of parasite at a time in the blood of malaria patients. Rest of merozoites act as male or female gametocytes. Again the merozoites enter in the digestive tract of new mosquitoes when they bite the infected person. The merozoites multiply in digestive tract of mosquito, pass through reproductive cycle and form infective sporozoites (Fig. 3.13).

Laboratory diagnosis of *Plasmodium* can be made by taking out blood samples, preparation of smear and microscopic observations for the presence of parasite. However, no effective immunity against *Plasmodium* develops besides some resistance in endemic areas.

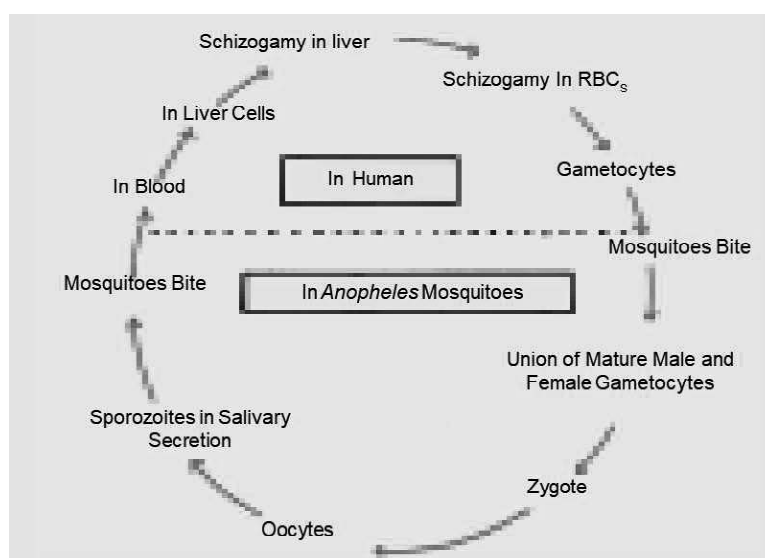


Fig. 3.13 Life cycle of *Plasmodium vivax*

B. Chemotherapy

Much researches have been done on chemotherapy of malaria. Quinine and derivatives of quinine e.g. chloroquine, primaquine, etc have been used for several years. Chloroquine inhibits DNA replication at merozoite stage. Now-a-days, resistance to these drugs have been reported. Therefore, in recent years drug combinations have been recommended e.g. Fansidar which is a combination of pyrimethamine and sulfadoxine. The combinations of drug act synergistically. However, effective control of malaria is not known except chemotherapy.

Malarial Vaccines

In recent years, much effort has been made for the preparation of effective vaccine through gene mapping, preparation of monoclonal antibodies, and protein synthesis. In 1975, Rath and Victor at New York University, Medical Care Centre, identified a surface antigen of the sporozoites known as circumsporozoite (CS). The CS antigen was used to produce the monoclonal antibodies against *Plasmodium*. In 1983, the CS gene was cloned to produce the CS protein in large quantities and to analyse and prepare antibodies (Miller *et al.*, 1986). In 1987, for the first time human test for vaccine was done at the university of Maryland, School of Medical Science (U.S.A.) and the effectiveness of vaccine was determined in 1989. But

much research work is needed on the preparation of vaccine for its use at several stages of the parasite.

Malaria Eradication Movement

Several times malaria eradication movement was launched by the government organizations to eradicate mosquitoes by spraying DDT, but the mosquitoes developed resistance. However, no significant control measure has been reported so far.

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2. *Balantidium coli*

Balantidium coli causes balantidiasis or balantidial dysentery. It is the only largest and ciliated pathogenic protozoan of human intestine (Fig. 3.14). *B. coli* enters in the intestine when its cysts present in contaminated food or water are ingested by humans. After ingestion the parasite reaches to the colon, wall of cysts dissolved and trophozoites are released. The trophozoites feed upon bacteria, faecal debris and tissue of colon. When the faeces get dehydrated and pass through colon, the parasite becomes encysted, discharged through faeces and contaminate the water and food. In colon, the parasite remains in two forms i.e. vegetative form (trophozoite) and cyst form. It lives in large intestine and rarely invades the epithelial lining and cause abdominal cramping, vomiting, weight loss. Due to invasion of epithelium, there develops ulcer in large and small intestines with total dysentery. The cysts can be observed upon microscopic observation of faeces. Life cycle of *B. coli* is shown in Fig. 24.5.

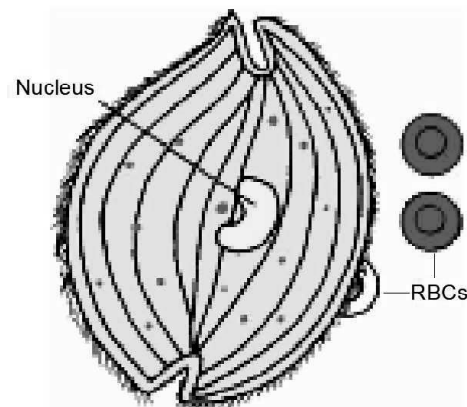


Fig. 3.14 Structure of *Balantidium coli*

3.. *Trichomonas vaginalis*

Trichomonas vaginalis is an inhabitant of vagina and urethra (Fig.24.6). It grows and increases its number over the normal microbial community, when acidity of vagina is disturbed from normal pH (3.8-4.4). The change in pH of vaginal fluid may be due to a loss of normal acid-producing bacterial flora or use of oral contraceptives. In such situation, it causes trichomoniasis or 'trich' in female and rarely in males. *T. vaginalis* is also transmitted by sexual intercourse or contaminated examination tools, towels, catheters, etc. After infection, it causes a mild inflammation of vagina, cervix and volva. It renders the dissolution of infected surface tissues. Consequently, a yellow or cream-white ooze of foul odour comes

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out. This type of disease is also caused by *Candida albicans* and *Gardnerella vaginalis*. In males, the prostate, seminal vesicle and urethra may be infected and consequently discharge a white liquid. *T. vaginalis* is observed under the microscope by taking the samples from the discharged fluid. The parasite is also found in semen or urine in males. Effective treatment is the use of oral metronidazole (flagyl), and the other antiprotozoan drugs.

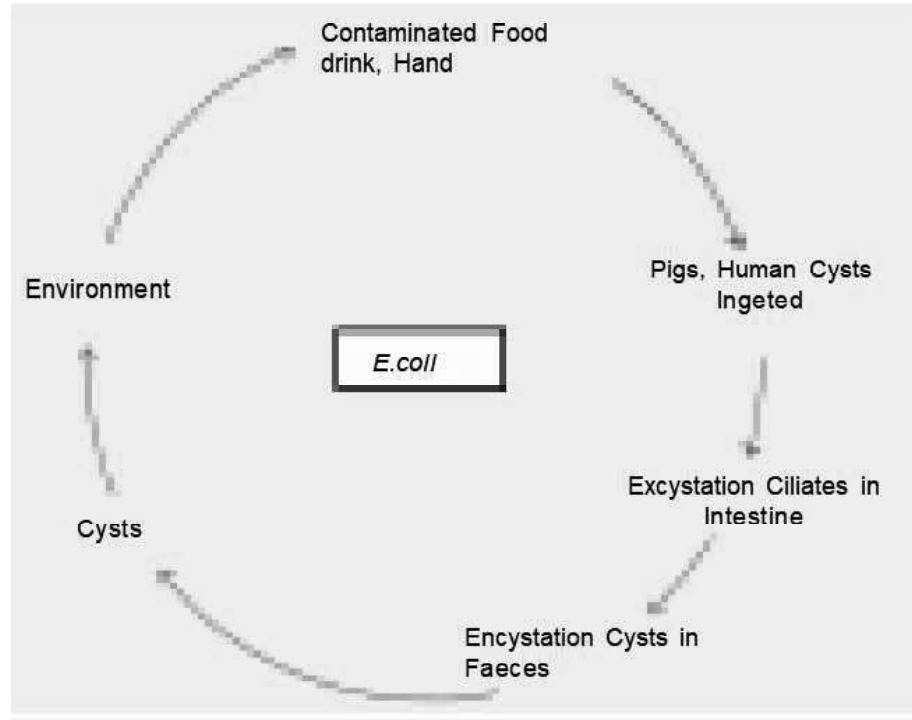


Fig. 3.15 Life Cycle of *Balantidium Coli*

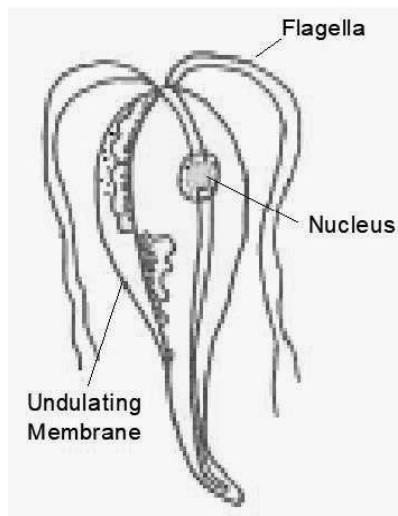


Fig. 3.16 Structure of *Trichomonas vaginalis* (trophozoite)

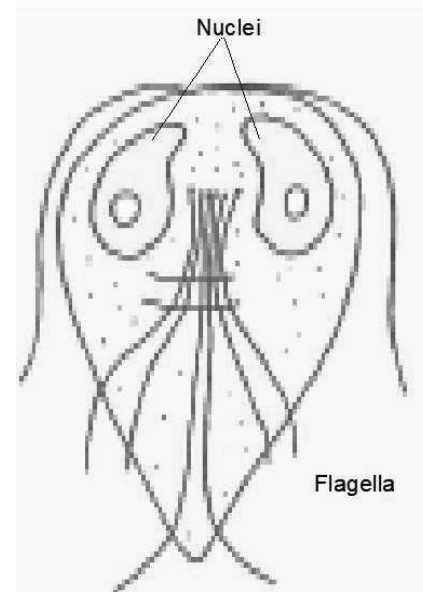


Fig. 3.17 Structure of *Giardia lamblia*

4. *Entamoeba histolytica*

The name of pathogen itself defines the disease (*ent*=inside, *amoeba*=varied shape, *histo*=tissue, *lytica*-bursting). It causes amoebiotic dysentery i.e. amoebiasis throughout the world. The parasite spreads through the contaminated water or food. The normal level of chlorine does not kill the amoeba. The cysts are not affected by the acid of stomach i.e. HCl. Only the vegetative cells are destroyed. On the epithelial cell of the wall of large intestine the vegetative cells multiply. This results in severe dysentery where the faeces contain *mucus* and blood. The life cycle of *E. histolytica* is given in Fig. 24.9. The pathogen feeds upon RBCs and damages the tissue of gastrointestinal tract. During severe cases, amoebae may enter the tissues of vital organs and cause abscess in liver, lungs, intestinal wall, etc. The intestinal wall becomes perforated. If abscess is formed it is treated surgically. The pathogen is identified by observing the amoebae in faeces and RBCs in trophozoites. Effective chemotherapeutic drugs are metronidazole plus iodoquinol.

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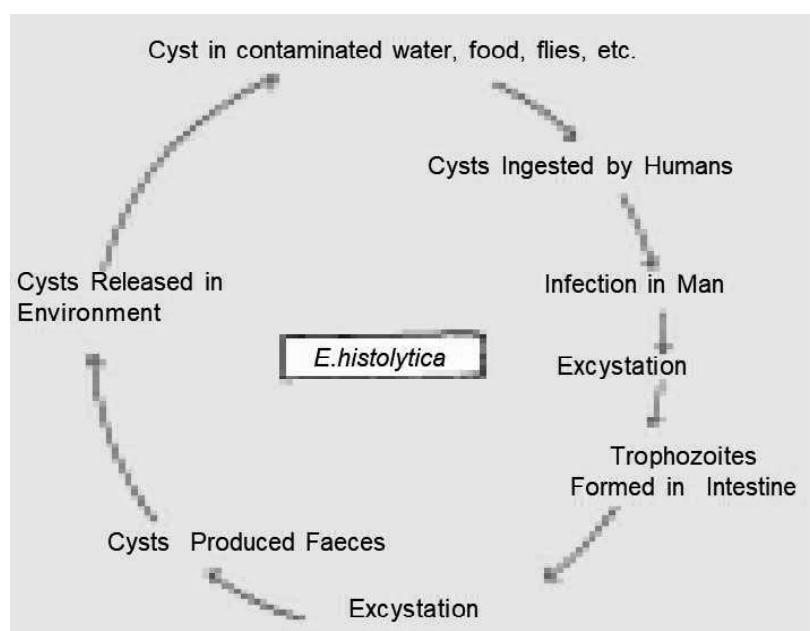


Fig. 3.18 Life cycle of *Entamoeba histolytica*

Check Your Progress

14. What are intestinal flagellates?
15. Name few diseases caused by *Trypanosoma*.
16. Which disease is caused by *Leishmania*?
17. Which is largest ciliated pathogenic protozoan of human intestine?
18. Why *Trypanosoma* is known as haemoflagellate?

3.5 ANSWERS TO 'CHECK YOUR PROGRESS'

NOTES

1. Parasites are living organisms that live in or on a host, which provides physical protection and nourishment.
2. Parasitic infections are more prevalent in warm climates, in less developed areas and among the socially deprived sections of society.
3. The parasites that inhabit the surface of the body of the host without penetrating into the tissues are called ectoparasites.
4. The indirect life cycle of a parasite is a situation in which a parasite requires two or more species of hosts to complete its life cycle.
5. The factors such as parasitic strain, number, size, metabolic process and host age, natural immunity, immunity to infection, co-existing diseases, nutritional status and life style have huge impact on the parasitic infection in man.
6. Plasmodium falciparum infection can cause blockage of capillaries of the cerebral cortex leading to fatal cerebral malaria.
7. A host is an organism that harbours a parasite.
8. The two kinds of parasitism are temporary parasitism and permanent parasitism.
9. Parasitology is the study of parasites.
10. Giardia and Cryptosporidium are two of the most common protozoa.
11. The stages in the life cycle of protozoa include:
 - Trophozoite
 - Merozoite
 - Gametocytes
 - Gametes
 - Oocysts
12. Protozoa respire by osmosis.
13. Endodyogeny is a method of reproduction in which the parasitic cell undergoes a single internal budding resulting in the formation of two daughter cells as seen in the genus toxoplasma.
14. Protozoa that use flagella as their mode of locomotion and reside in the intestine of the host are called intestinal flagellates.
15. Sleeping sickness (African trypanosomiasis) and Chagas disease (American trypanosomiasis) are caused by different species of Trypanosoma.
16. Leishmania is a genus of trypanosomatid protozoa. It is the parasite that is responsible for the disease leishmaniasis.
17. Balantidium coli is the only largest ciliated pathogenic protozoan of human intestine.
18. Trypanosoma is known as haemoflagellate as it infects the blood.

3.6 SUMMARY

- Parasites are living organisms that live in or on a host, which provides physical protection and nourishment.
- Host is an organism, which harbours the parasites. The host offers some resistance to the injury done by the parasite and there may be some adaptation or tolerance between the parasite and the host.
- Anthroponoses is a parasitic infection that is confined to humans, for example, malaria and filaria.
- Definitive host is a kind of host in which the parasite spends the adult stage of its life or utilizes a sexual method of reproduction. In most human parasitic infections, man is the definitive host; however, man acts as intermediate host in malaria and hydatid disease.
- Intermediate host is a kind of host in which the larval stage of the parasite either lives or the asexual multiplication takes place.
- Sometimes, larval developments are completed in two different intermediate hosts called first and second intermediate hosts respectively.
- Paratenic host is kind of host in which a parasite merely remains viable without any further development. Such host can transmit the infection to another host.
- Such hosts are also called a transport hosts or mechanical hosts, e.g., a fly may transfer amoebic cysts from infected faeces to food that is eaten by humans.
- Reservoir host is a kind of host that sustains a parasitic infection in an endemic area.
- A reservoir host is an animal or man in which a parasite commonly resides or one in which a parasite that infects man is able to be maintained in the absence of a human host.
- In majority of parasitic infections, man is the main reservoir.
- In some cases, both man and animals are infected and the latter serves as reservoirs of infection.
- The clinical manifestations depend upon the number of invading organisms enter during primary infection and reinfection.
- Just like in viral infections, the anti-parasitic antibody is only effective against extracellular parasites and if parasitic antigens are exhibited by the infected cells on their surfaces.
- Neutralization occurs when the antibody combines with various surface molecules of parasites and then neutralizes them, blocking or interfering with their proper functioning.
- Antibody binds to toxins and enzymes of certain parasites. This protects the host from damage. It also affects the infection process.

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- Parasites can enter the host's immune responses by changing their antigenic structure, Intracellular location and change of the host response to favour the survival of the parasite.
- The molecular methods of grouping organisms are important in confirming taxonomic conclusions and as studies continue in parasitic genetics, immunology and biochemistry, the species designation will become more defined.
- All protozoa can be transformed from an active or trophozoite stage to an inactive (or resting) or cystic stage that encloses itself within a tough wall.
- Infection with *Giardia lamblia* may result into asymptomatic to severe giardiasis.
- Trypanosomes are meta-cyclic and are transmitted by inoculation through proboscis of the insect vector.
- *Leishmania* is a genus of trypanosomatid protozoa. It is the parasite that is responsible for the disease leishmaniasis.
- Toxoplasmosis is caused by the protozoan parasite *Toxoplasma gondii*.
- There are many free-living amoebae, but only few are known to cause infections in humans, e.g., *Naegleria fowleri*, *Acanthamoeba* spp., *Ballamuthia mandrillaris* and *Sappinia pedata*.

3.7 KEY TERMS

- **Parasites:** These are living organisms that live in or on a host, which provides physical protection and nourishment.
- **Medical parasitology:** It deals with parasites, which infect man and the diseases produced by them.
- **Host:** It is an organism, which harbours the parasites. The host offers some resistance to the injury done by the parasite and there may be some adaptation or tolerance between the parasite and the host.
- **Symbiosis:** It is an association in which both parasite and host are so dependent upon each other that they cannot live without the help of the other, and both the partners do not face any harm from the association.
- **Commensalism:** It is an association in which the parasite derives benefit without causing any harm to its host.
- **Ectoparasites:** These parasites inhabit the surface of the host's body without penetrating into the tissues.
- **Endoparasites:** These parasites live within the host's body in the blood, tissues, body cavities, digestive tract and other organs.
- **Definitive host:** It is a kind of host in which the parasite spends the adult stage of its life or utilizes a sexual method of reproduction
- **Intermediate host:** It is a kind of host in which the larval stage of the parasite either lives or the asexual multiplication takes place.

- **Paratenic host:** It is kind of host in which a parasite merely remains viable without any further development.
- **Reservoir host:** It is a kind of host that sustains a parasitic infection in an endemic area. A reservoir host is an animal or man in which a parasite commonly resides or one in which a parasite that infects man is able to be maintained in the absence of a human host.
- **Binary fission:** It is the most common method of multiplication and it occurs by mitotic division of the nucleus that is followed by division of the cytoplasm.
- **Intestinal parasites:** These are parasites that thrive within the intestines of other organisms, including humans, for years without causing any symptoms.
- **Protozoa:** These are a subcategory of parasites that are single-celled organisms and cause infections and diseases when they come in contact with humans.
- **Trophozoite:** It is a generic term for the moving, feeding and reproducing stages of most protozoa.

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3.8 SELF-ASSESSMENT QUESTIONS AND EXERCISES

Short-Answer Questions

1. What are the two types of life cycles of parasites?
2. Write the mechanisms of transmission of parasitic infections.
3. Name some diseases caused by protozoa?
4. How the giardiasis can be diagnosed?
5. State the various stages of the life cycle of Leishmania.
6. Write the morphology of Toxoplasma.
7. Which is the causative organism of malaria in humans?
8. What do you know about malarial vaccines?

Long-Answer Questions

1. Explain the different types of hosts in cycle of parasites.
2. Analyze the cellular response in parasitic infection.
3. Describe the general characteristics of protozoa.
4. Explain the clinical manifestation of Giardia intestinalis.
5. Briefly describe the laboratory diagnosis of Leishmania infections.
6. Explain the pathogenesis and risk factors of Toxoplasmosis.
7. Describe the life cycle of Plasmodium vivax.

3.9 FURTHER READING

NOTES

- Pawar, Kishore R, Ashok E Desai. 2016. *Parasitology*. New Delhi: Nirali Prakashan.
- Heelan, Judith Stephenson, Frances W. Ingersoll. 2002. *Essentials of Human Parasitology*. US: Delmar publishers
- Sood, Ramnik.2019. *Textbook of Human Parasitology Protozoology and Helminthology*. New Delhi: CBS PUB & DIST PVT Limited INDIA.
- Myerowitz, Richard L. 1983. *Zygomycosis (mucormycosis) The Pathology of Opportunistic Infections with Pathogenetic, Diagnostic, and Clinical Correlations*. New York: Raven Press.

UNIT 4 BIOLOGY OF PARASITISM II

Structure

- 4.0 Introduction
- 4.1 Objectives
- 4.2 Pathogenicity of Helminths
 - 4.2.1 Plathyleminthes Parasites
 - 4.2.2 Nematelminthes Parasites
- 4.3 Ectoparasites
- 4.4 Parasitic Adaptations in Parasites
- 4.5 Evolution of Parasitism
- 4.6 Answers to ‘Check Your Progress’
- 4.7 Summary
- 4.8 Key Terms
- 4.9 Self-Assessment Questions and Exercises
- 4.10 Further Reading

NOTES

4.0 INTRODUCTION

Parasitism is a kind of symbiosis, a close and persistent long-term biological interaction between a parasite and its host. Unlike saprotrophs, parasites feed on living hosts, though some parasitic fungi, for instance, may continue to feed on hosts they have killed. Unlike commensalism and mutualism, the parasitic relationship harms the host, either feeding on it or, as in the case of intestinal parasites, consuming some of its food. Because parasites interact with other species, they can readily act as vectors of pathogens, causing disease. Parasites may be characterized as ectoparasites—including ticks, fleas, leeches, and lice which live on the body surface of the host and do not themselves commonly cause disease in the host; or endoparasites, which may be either intercellular (inhabiting spaces in the host’s body) or intracellular (inhabiting cells in the host’s body). Intracellular parasites—such as bacteria or viruses—often rely on a third organism, known as the carrier, or vector, to transmit them to the host.

Helminths are worm-like parasites that survive by feeding on a living host to gain nourishment and protection, sometimes resulting in illness of the host. There are a variety of different helminths from the very large to the microscopic. Parasitic worms live in and feed in living hosts. They receive nourishment and protection while disrupting their hosts’ ability to absorb nutrients. This can cause weakness and disease in the host, and poses a global health and economic problem. Parasitic worms cannot reproduce entirely within their host’s body; they have a life cycle that includes some stages that need to take place outside of the host. Helminths are able to survive in their mammalian hosts for many years due to their ability to manipulate the host’s immune response by secreting immunomodulatory products. All parasitic worms produce eggs during reproduction. These eggs have a strong shell that protects them against a range of environmental conditions. The eggs can therefore survive in the environment for many months or years.

In this unit you will study about plathyleminthes parasites - pathogenic to man, nemathelminthes parasites, ectoparasites, vectors of human diseases, parasitic adaptations in parasites, and evolution of parasitism.

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4.1 OBJECTIVES

After going through this unit you will be able to:

- Explain plathyleminthes parasites
- Define nemathelminthes parasites
- Understand ectoparasites
- Analyze vectors of human diseases
- Elaborate on parasitic adaptations in parasites
- Explain evolution of parasitism

4.2 PATHOGENICITY OF HELMINTHS

The majority of metazoan parasites of vertebrates are representatives of two phyla, which are as follows:

- (i) Acoelomate platyhelminths
- (ii) Pseudocoelomate nematoda

These parasites are usually referred to by the word, ‘helminths’ and this term is made to include all the cestodes and digeneans of the former group and all members of the latter. Of the four classes of entirely parasitic platyhelminths, cestodes and digeneans are medically important in humans. Helminths also cause debilitating, deforming and fatal diseases in humans as shown in Tables 4.1, 4.2 and 4.3.

Table 4.1 Helminths in Humans

| Phylum Platyhelminthes (flatworms) | Phylum Nematoda (roundworms) |
|---|---|
| Monogenea | Rhabditida (<i>Strongyloides</i>) |
| Cestoda (<i>Diphyllobothrium, Taenia, Echinococcus</i>) | Strongylida (<i>Necator, Ancylostoma, etc.</i>) |
| Aspidogastrea | Ascaridida (<i>Ascaris, Toxocara, etc.</i>) |
| Digenea (<i>Schistosoma, Fasciolopsis, Fasciola, Paragonimus</i>) | Oxyurida (<i>Enterobius</i>) |
| | Spirurida (<i>Dracunculus, Wuchereria, Brugia, Loa, Onchocerca</i>) |
| | Enoplida (<i>Trichinella, Trichuris</i>) |

Table 4.2 Larvae Helminths in Humans

| Flukes (Trematodes) | Tapeworms (Cestods) | Roundworms (Nematodes) |
|---------------------|-------------------------------------|------------------------|
| Miracidium | Cysticercus | Rhabditiform |
| Sporocyst | Cysticercoid | Filariform |
| Redia | Coenurus Coracidium | Microfilaria |
| Cercaria | Proceroid | |
| Metacercaria | Plerocercoid (sparganum) Hydatid | |

Table 4.3 Helminth Stages Changing Pathology in Humans

| Helminths | Egg | Larvae | Adult |
|-----------|-----|--------|-------|
| Flukes | + | + | + |
| Tapeworms | – | + | + |
| Nematodes | – | + | + |

NOTES

The word ‘helminth’ has been derived from the Greek word ‘helmins’ meaning worms. The helminthic parasites are multicellular and bilaterally symmetrical in appearance. They possess three germinal layers and are therefore called triploblastic metazoan. The term ‘helminth’ was originally used with reference to intestinal worms. However, now it includes both the tissue worms as well as many free-living parasites. The nematode species of helminths indirectly infect humans causing diseases. The larvae of these parasites cause serious zoonotic diseases (diseases transmitted from animals to humans). During their life cycle, these parasites can and do infect many intermediate hosts and normally, the animals are definitive hosts while humans are accidental definitive hosts.

Nematodes

Filariae consist of a group of nematodes that invade the blood stream, connective tissue or serous cavities of vertebrates. Morphologically, they are long thread-like nematodes. Many of them are medically important as they attack humans and are transported by various vectors like mosquitoes. The filarial nematodes that can parasitize humans include the following:

- *Wuchereria bancrofti*
- *Brugia malayi*
- *Brugia timori*
- *Loa loa*
- *Onchocerca volvulus*
- *Mansonella perstans*
- *Mansonella streptocerca*
- *Dipetalonema streptocerca*

These parasites can inhabit lymph glands, deep connective tissues, subcutaneous tissues or mesenteries and result in inflammatory reactions or fleshy deformities known as elephantiasis. The nematodes that are important in humans are the following:

I. Tissue nematodes

- *Onchocerca volvulus*
- *Toxocara canis*
- *Trichinella spiralis*
- *Dracunculus medinensis*

II. Blood nematodes

- *Wuchereria bancrofti*
- *Loa loa*
- *Brugia malayi/timori*

NOTES

The general characteristics of nematodes are as follows:

- They belong to the phylum nematoda.
- They are non-segmented, cylindrical worms that taper at both ends.
- They have a shiny, tough, cellular, hyaline cuticle.
- They are dioecious and the male is smaller than the female and the posterior end of the former is curved.
- The female worms possess one or two tubular ovaries which lead to a uterus or united uteri. They are viviparous, oviparous or ovoviviparous.
- They also moult, which is a feature in the development of nematode larvae.

Helminths develop in several stages—egg and larval (juvenile) to the adult stage. Platyhelminths and nematodes that can infect humans share similar anatomic features. They have common physiologic requirements and functions and their outer covering is known as a cuticle or tegument. They also have prominent external structures of flukes and cestodes are acetabula (suckers) or bothria (false suckers). The male nematodes of several species have accessory clear sex organs which are basically modifications of the cuticle. The alimentary, excretory and reproductive systems can also be clearly identified in them. However, the tapeworms do not have an alimentary canal and the nutrients are absorbed through the tegument. Both the blood flukes and nematodes are bisexual.

All other flukes and tapeworm species that infect humans are hermaphroditic. The flukes, cestodes and nematodes produce eggs and though the sexes are different, the structure of the body is the same as that of hermaphroditic flukes. The male and female worms inhabit the lumen of blood vessels. All medically important flukes are of the taxonomic category. Digenea (with a developmental cycle) needs at least two hosts.

The eggs that are passed by the faeces, urine or sputum of humans reach an aquatic condition where the eggs hatch and release ciliated larvae or miracidia that either penetrate or are eaten by an intermediate host. After this, a sporocyst or redia stage develops from a miracidium in the host that provides rediae or a daughter sporocyst stage. It is in this stage where cercariae develop asexually, in order to migrate to an aquatic condition. They either penetrate the definitive host (to become adults) or a second intermediate host (to develop as an encysted metacercariae). Finally, after a metacercarial cyst is ingested, the digestion of the cyst produces an immature fluke that migrates to a specific organ site, which ultimately develops into an adult worm (Table 4.4).

Table 4.4 Differences between Pseudophyllidean and Cyclophyllidean Tapeworms

| Differentiating Feature | Differentiating Feature | Differentiating Feature |
|-------------------------|---|--|
| Scolex | Two sucking grooves (bothrial) | Four muscular suckers (acelabula) |
| Genital pore | Center of each proglotid | Margni(s) of each proglottid [may be located On both sides in an irregular pattern (<i>Taenia</i> spp); all on the same side (<i>Hymenolepis</i> spp); or each proglotid may have a pore on each side (<i>dipylidium caninum</i>)] |
| Uterine pore | Center of proglottides on ventral surface | Absent; uterus ends blindly |
| Uterus (gravid) | Relatively long and coiled | Saclike, highly branched |
| Egg | Operculate | Nonoperculate |
| Oncosphere | Ciliated (coracidium) | Nonciliated |
| Larvae | Proceroid and plerocecold; both forms solid | Cysticercold, cysticecus, hydatid; all forms cystic |

NOTES

Cestodes

Cestodes are dorsoventral worms that are commonly known as tapeworms. They possess a segmented body that resembles a measuring tape and the term ‘tapeworm’ is thus used. Their length varies from a few millimeters to several meters. Various adult tapeworms that infect humans have been named after their intermediate host—the fish tapeworm (*Diphyllobothrium latum*) and the pork tapeworm (*Taenia solium*). The medically important species of tapeworms include the following:

- *Taenia saginata*
- *Taenia solium*
- *Echinococcus granulosus*
- *Hymenolepis nana*

Trematodes

In medical terms, the most important trematodes are blood flukes (*Schistosoma mansoni*, *S. japonicum* and *S. hematobium*), the intestinal fluke (*Fasciolopsis buski*), the liver fluke (*Clonorchis sinensis*) and lung fluke. The worms are usually are 10–20 mm in length and the male has an unusual lamelliform shape with marginal folds that form a canal in which the slender female worm resides. The trematode parasites of humans and animals are as follows:

- *Fasciolopsis buski* (intestinal fluke)
- *Fasciola hepatica* (sheep liver fluke)
- *Clonorchis sinensis* (human liver fluke)
- *Paragonimus westermani* (lung fluke)
- *Paragonimus mexicanus* (Mexican lung fluke)
- *Schistosoma mansoni* (blood fluke)
- *Schistosoma haematobium* (blood fluke)
- *Schistosoma japonicum* (blood fluke)
- *Avian schistosomes* (Avian blood fluke)

4.2.1 Plathyleminthes Parasites

NOTES

Taenia saginata, also known as the beef tapeworm, is both a cattle and a human parasite. It occurs when the cattle are raised by the infected humans who maintain poor hygiene, do not properly dispose human faeces, have poor meat inspection programmes and eat meat without properly cooking it. This disease is relatively common in Africa, Eastern Europe, Southeast Asia and Latin America.

Morphology of *T. saginata*

Taenia saginata is a worm that is 4–6 meters long and 12 mm broad. Its head is pearshaped and it has four suckers but no hooks or neck. Its body is long and flat. Its segments are made up of mature and gravid proglottids. The size of the segments is approximately 18×6 mm. It has a branched uterus and its egg is yellow-brown and 35×45 micrometers in size. The egg is round and has peripheral radial striations and it contains an embryo that has three hooklets.

Some important points about *T. saginata* are as follows:

- *T. saginata* lives in human intestine and has a life span of more than 10 years.
- *T. saginata* has four cup shaped suckers.
- The strobila of *T. saginata* has 1000–2000 proglottids.
- The gravid segments in *T. saginata* are capable of movement and they migrate to the grazing sites where they rupture to release the eggs.
- Gravid segments of *T. saginata* migrate through the anus and then they rupture to release the eggs.
- The eggs of *T. saginata* are spherical in shape.
- The larval form of *T. saginata* is called *Cysticercus bovis*.

Life Cycle of *T. saginata*

Life cycle of *T. saginata* goes through many stages. Some of the important points about those stages have been discussed as follows:

- Humans act as the definitive host and cattle act as the intermediate hosts for *T. saginata*.
- The infective form for human is known as *C. bovis*.
- The larvae of *T. saginata* are present in the muscles of cattle and the infection in human occurs by the consumption of raw or undercooked beef that contains cysticerci.
- The worm invaginates in the small intestine and gets attached to the mucosa by suckers.
- Eggs are formed in the gravid segment of the worm that ruptures in the intestine on the perianal skin.
- While grazing, the cattle ingests the eggs along with grass and these eggs hatch and penetrate the mucosa to invade the bloodstream or lymphatics to reach the muscles.

- The oncosphere metamorphoses into *C. bovis* in a span of two and half months.
- Cysticercus develops in striated muscles of hind limbs, diaphragm and tongue.
- When the beef infected with *C. bovis* is eaten in a raw or undercooked form by humans the life cycle of *T. saginata* gets completed.

Figure 4.1 shows the life cycle of *T. saginata*.

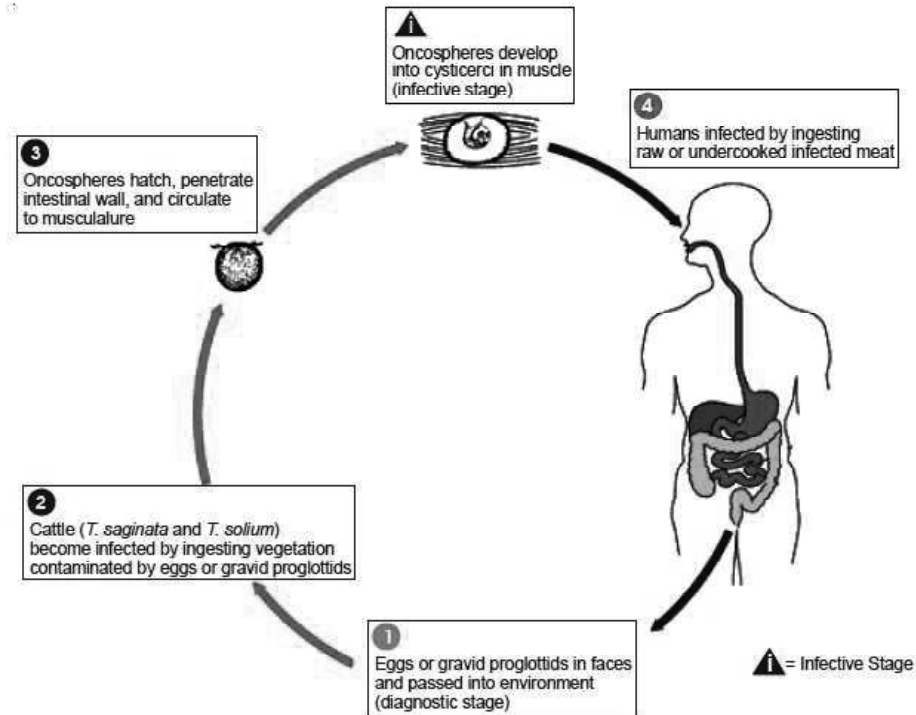


Fig. 4.1 Life Cycle of *T. saginata*

Clinical Picture

- Adult worms give abdominal discomfort and bowel disturbances.
- Cysticercosis produces palpable nodule in muscles
- Neurological manifestations like seizures and focal neurological deficits may develop because of inflammation. Hydrocephalus and raised intra-cranial pressure may result.
- Calcified cysticerci are seen in soft tissue or brain.

Laboratory Diagnosis of *T. saginata*

Laboratory diagnosis of *T. saginata* is done as follows:

- Taenia eggs and proglottids are identified through microscopic identification. However, this technique is not possible during the first 3 months following infection, prior to the development of adult tapeworms.
- The species can also be distinguished by examining proglottids. Three consecutive day examination of faeces detects *T. saginata* eggs in upto 80% patients.

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- The immunological tests can also detect the presence of cysticerci and improved imaging techniques such as CAT and MRI can be very useful in detecting cysticerci in various organs.
- Diagnosis can also be made in the perianal swab where the eggs that are deposited in the perianal region by the motile proglottids, are detected microscopically in 85 to 95% patients.
- A definitive diagnosis can be established by demonstration of an unarmed scolex after anti-helminthic treatment.
- CSF studies examination shows raised proteins, reduced sugar and pleocytosis.
- ELISA on CSF or sera to detect antibodies against purified antigens of cysticerci establishes the diagnoses.

T. Saginata and T. solium

T. solium, also called the pork tapeworm, is a cyclophyllid cestode in the family Taeniidae. It infects pigs and humans in Asia, Africa, South America, parts of Southern Europe, and pockets of North America. The adult tapeworms have an average length of ~3 meters that can go up to 8 meters. They contain between 800 and 1000 proglottids. The gravid proglottid is 12 mm long.

Some of the important points about *T. solium* and *T. saginata* are as follows:

- *T. solium* is smaller than *T. saginata*.
- *T. solium* has a globular scolex with four suckers and a circular row of hooks, which imparts a solar appearance.
- Eggs of *T. solium* and *T. saginata* cannot be distinguished.
- *T. saginata* has 15 to 20 branches on each side, while *T. solium* has 7 to 13.
- Unlike *T. saginata*, the scolex in *T. solium* is equipped with a double crown at approximately 30 hooks.

Life cycle of T. solium

The various stages of the life cycle of *T. solium* are as follows:

- The definitive host is human and the intermediate host is pig. Man ingests the larvae by eating uncooked contaminated pork that possesses the larvae in cysticerci.
- The larvae develop into adult form called the tapeworm.
- Tapeworm attaches to the intestinal lining of human and sheds eggs in human feces.
- Pigs come into contact with human feces. They ingest the tapeworm eggs.
- The eggs penetrate the small intestine of the pig; enter the hepatic portal vein to enter the general circulation.
- The eggs migrate to the skeletal or cardiac muscle. Here they form cysticerci.

Sometimes an infected human may ingest the eggs found in its feces and cysticerci may develop throughout the tissues of the body, particularly the brain. This gives rise to Neurocysticercosis.

Figure 4.2 shows the life cycle of *T. solium*.

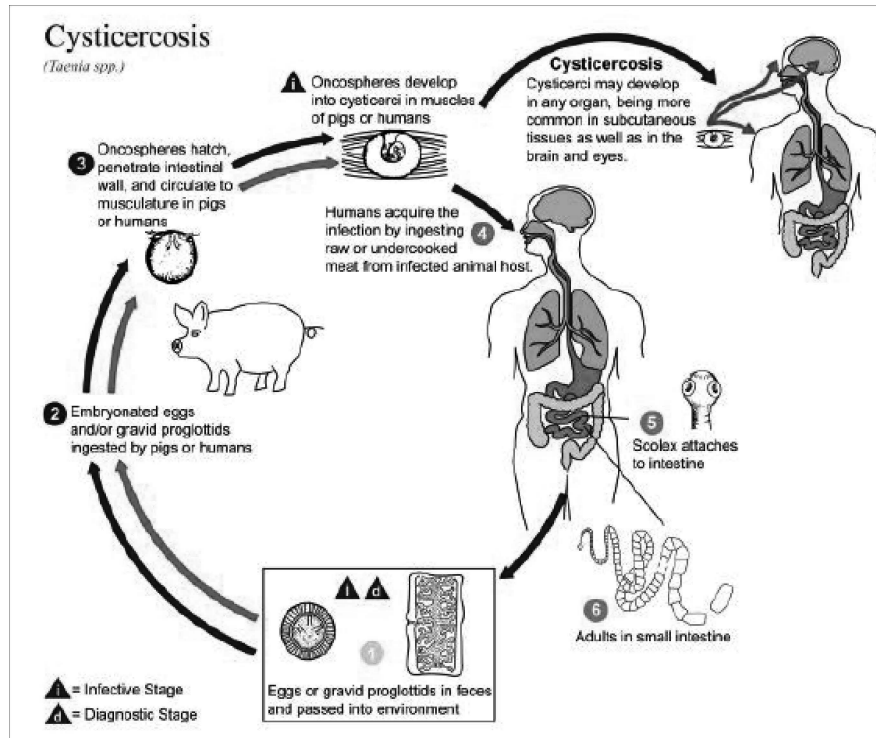


Fig. 4.2 Life Cycle of T. solium

Intestinal Helminths

Intestinal helminths are a type of intestinal parasite that resides in the human gastrointestinal tract. Because of their high mobility and lower standards of hygiene, school-age children are particularly vulnerable to these parasites.

Helminths can live within the intestines for years without causing any symptoms. When they do, symptoms include the following:

- Abdominal pain
- Diarrhea
- Nausea or vomiting
- Gas or bloating
- Dysentery (loose stools containing blood and mucus)
- Rash or itching around the rectum or vulva
- Stomach pain or tenderness
- Feeling tired
- Weight loss
- Passing a worm in your stool

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Guidelines for Identification of Intestinal Helminths

Intestinal helminths can be identified by fecal testing. Stool samples must be collected before you take any anti-diarrhea drugs or antibiotics. During examination, the following guidelines must be followed:

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- Eggs must be examined for size. The length and width must be measured and they must fall within a specific range.
- The shape of the egg must be taken into account as each species has its own particular shape.
- The stage of development must be recorded.
- The number of cells present in the eggs must be identified. This is necessary because while some species have single celled eggs, others have multi-celled eggs.
- It must be checked whether the eggs have embryonated in feces or not as the eggs of some species do so.
- The thickness of the egg shell must also be identified.

4.2.2 Nemathelminthes Parasites

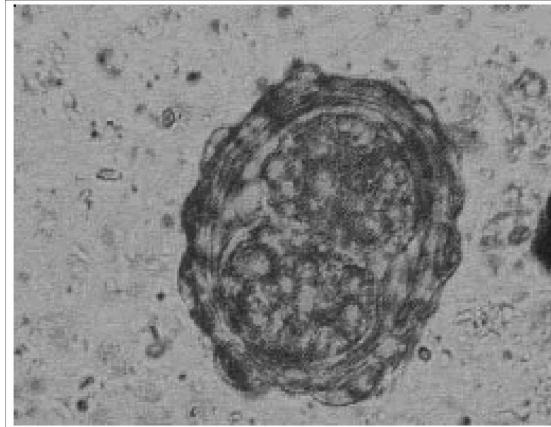
Ascaris lumbricoides is commonly called ‘round worm.’ It is the biggest intestinal nematode that infects humans. It is found worldwide and is most prevalent throughout the tropics and sub-tropics.

In terms of epidemiology, this infection has worldwide distribution and there are many conditions that favour its transmission to humans. Some of these conditions are as follows:

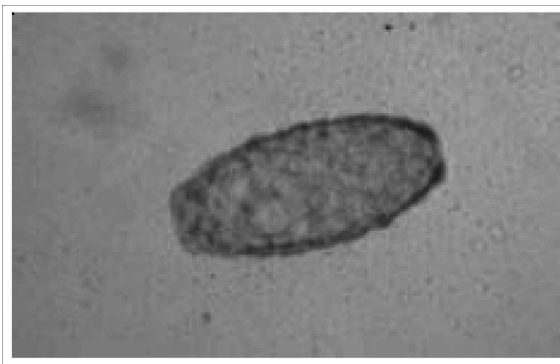
- Adoption of a simple life cycle
- Production of up to 240,000 eggs/day and these eggs being highly resistant to the routinely used sanitizers because of the presence of ascroside
- Specific social customs and living habits.

Morphology of *A. lumbricoides*

An adult worm is cylindrical in shape and creamy white to pinkish in colour. On an average a female worm size ranges from 20–35cm in length whereas a male worm is smaller in size. Its average length is 15–31cm and it is slimmer than a female worm. A tapeworm has a curled tail that looks like a pair of sickle copulatory spines. It has a completely different digestive tract and its reproductive organs are tubular in shape. Usually, a male has a single reproductive tubule whereas a female tapeworm has two reproductive tubules and the vulva is ventrally located at the posterior part of the anterior body. Figure 4.3 (a) and (b) show fertilized and unfertilized ovum of *Ascaris lumbricoides*.



(a) Fertilized Ovum of *Ascaris lumbricoides*



(b) Unfertilized Ovum of *Ascaris lumbricoides*

Fig. 4.3 Morphology of *Ascaris Lumbricoides*

Life Cycle of *A. lumbricoides*

Ascaris lumbricoides inhabits the small intestine and the stage when it infects is the stage in which the eggs are embryonated. The infection generally spreads from the mouth and it does not have any intermediate and reservoir hosts. An adult worm's life span is one year. It generally lives in the small intestine's lumen and the intestinal content is its food. A fertilized female lays its eggs here. Every female has the capacity to produce about 240,000 eggs every day and these eggs generally pass in the faeces. When they are passed, the eggs become unsegmented, but they still need about three weeks of outside development before a motile embryo gets formed within an egg.

Blood-lung migration phase of larvae: The larvae might cause pneumonia during their migration through the lungs. Pneumonia symptoms are as follows:

- Low fever
- Cough
- Blood-tinged sputum
- Asthma

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A lot of allergic systems also occur, generally, even leading to eosinophilia. These clinical manifestations lead to Loeffler's syndrome.

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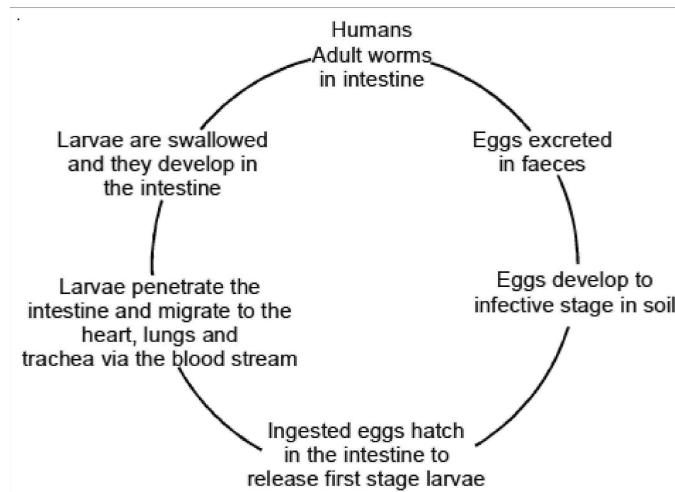


Fig. 4.4 Ascaris Lumbricoides

Laboratory Diagnosis of *A. lumbricoides*

A true and complete diagnosis of *A. lumbricoides* is dependent on the worm or the egg's recovery and identification.

- **Ascaris Pneumonitis:** It is the examination of sputum for *Ascaris* larvae.
- **Intestinal Ascaris:** It is the examination of faeces for the *Ascaris* eggs. There are various methods of doing it. Some of them are as follows:
 - o Direct fecal film
 - o Brine-floatation method
 - o Recovery of adult worms

The microscopic examination of stool deposits after concentration gives the characteristic bile stained ova. Unfertilized ova are brick shaped that give irregular bumpy surface and measure, 85–95µm by 43–47µm. The fertilized ova are oval shaped with a thick walled and irregular bumpy surface. They measure 45–75µm by 35–50µm. The eggs are difficult to be identified and if an excess of iodine is added to the wet mount preparation, they retain the stain, thus resembling debris.

Check Your Progress

1. Name the major two phyla of metazoan parasites of vertebrates.
2. List some of the filarial nematodes that can parasitize humans.
3. What are cestodes?
4. At which geographical locations *Taenia saginata* is usually found?
5. How many cup-shaped suckers does *Taenia saginata* have?
6. What does the CSF examination of *Taenia saginata* show?

4.3 ECTOPARASITES

Ectoparasites - pathogenic to man. as vectors of human diseases

Ecto is a prefix meaning 'outer'. Ectoparasites refers to the organisms that are found either on the skin or only in the superficial layers of the skin. Almost, all ectoparasites are arthropods; i.e., they are invertebrates with a chitinous exoskeleton. The ectoparasites that cause human disease fall into two main categories: insects (six-legged arthropods) and arachnids (eight-legged arthropods).

In this section, we shall discuss insects like lice, fleas and bed-bugs and arachnids such as mites, ticks, and spiders. Several arthropods are vectors that transmit the organisms that cause important infectious diseases. A 'vector' refers to the organism which can pass on the disease to other organism without actually getting affected by that disease. Insect acts as vector of disease in two ways:

- (a) **Mechanical transmission-** The disease-causing pathogens are carried along with hairs, legs, antennae etc. of the insect. The infectious agent is passed on from one host to other without undergoing a stage of development inside the vector. This is referred to as mechanical transmission of the infectious agent by the vector.
- (b) **Biological transmission-** The disease-causing pathogens are carried inside the system of the insect. The infectious agent undergoes development or multiplication inside the vector. This is referred to as biological transmission of the infectious agent by the vector.

Before, moving further, let us study about the key identification features of class Insecta and Arachnida.

Body of Insect

The body is divided into three distinct regions: head, thorax and abdomen.

(a) Head: Head contains:

- One pair of antennae:
 - o The antennae are usually used as tactile organs (= organs pertaining to the sense of touch)
 - o They can also be used as olfactory organs (= organs of smell).
- Eyes: Most insects possess one pair of compound eyes and sometimes some simple eyes called "ocelli".
- Mouthparts: There is a big variety in types of mouthparts; biting, sucking, stinging, licking, etc.

(b) Thorax: Thoracic region contains:

- Three pairs of legs.
- The thorax has three segments.
- These are known as pro-thorax, meso-thorax and meta-thorax.
- Each segment has one pair of legs.

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- The different parts of the leg are called coxa, trochanter, femur, tibia, and tarsus.
- Some insects are legless, or have fewer than 6 legs.
- Some larvae have leg-like appendages on the abdomen.
- Often one or two pairs of wings.
- The wings are borne by the second and/or third of the thoracic segments.

Note: Some insects are wingless.

(c) Abdomen:

- The gonopore (genital opening) is at the posterior end of the abdomen.
- No appendages used for moving on the abdomen of adults (except in a few primitive insects).
- Sometimes there are some appendages at the end of the abdomen.

| Class Insecta |
|---|
| <i>Identification</i> 1. Three distinct body regions (head, thorax, abdomen) 2. Three pairs of legs 3. Often have wings 4. One pair of antennae 5. Segmented abdomen |
| Order Diptera ■ Mosquitoes ■ Flies |
| Order Heteroptera ■ Bugs |
| Order Anoplura ■ Lice |
| Order Siphonaptera ■ Fleas |
| Order Dictyoptera ■ Cockroaches |

Fig.4.5 Identification Features Of Members of Class Insecta

Figure 4.5 depicting the key identification features of members of class Insecta along with order to which the ectoparasites belong.

Arachnids are a class (Arachnida) of joint-legged Invertebrate Animals in the subphylum Chelicerata. All Arachnids have multiple legs and look like Insects, the only difference is that Insects have six leg and Arachnids have 8 legs. They are largely terrestrial and solitary animal, gathering only for mating. Fossils suggest

that Arachnids were among the first animals to live on land, perhaps in the early Devonian Period, nearly 400 million years ago. Some of the common Arachnids are Spiders, Scorpions, Ticks and Mites. Ticks and Mites belongs to order Acarina.

(1) Ectoparasite Insects

(a) **Head Louse (*Pediculus humanus*) as Ectoparasite**

- Human lice are ubiquitous, i.e., widespread. The habitat of the human louse is solely on the human body or in the clothes. They fail to survive anywhere else as they cannot survive away from the host for a few days.
- Human lice are more prevalent in unhygienic areas where people change or wash their clothing infrequently and/or are unclean themselves (Milne and Milne 1980).
- Lice are obligate ectoparasites; they feed on blood of human.
- They possess especially designed mouth parts for piercing the skin of humans as well as for retrieving the blood.
- *Pediculus humanus* is a small insect having a large abdomen.
- The legs of the *Pediculus humanus* are equipped with sharp claws for holding onto hair and clothing fibres.



Fig. 4.6 *The Pediculus Humanus*

Figure 4.6 is depicting the *Pediculus humanus*

- The head of the head louse is somewhat narrower than the body.
- They lack wings like most insects.
- They possess piercing mouthparts for digging into the skin and draining out the blood.
- *Pediculus humanus capitis*, is approximately 1-2 mm long, whereas the body louse, *Pediculus humanus humanus*, is usually slightly larger, having a size of 2-3.5 mm.
- The ‘nits,’ or eggs, of the head louse are approximately 1 mm long and about half as wide.
- The young lice are often referred to as ‘red backs’, as they become red in colour due to feeding blood.
- They turn to a grey colour, after the blood is digested.

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- The female lice lay their eggs, which are referred to as nits, singly on the hairs of the host (if they are head lice) or attached to clothing in case of body lice.
- The nits will hatch into nymphs in a time span of eight days.
- The nymphs also suck blood and mature in eight to sixteen days.
- Each adult female body louse produces between two and three hundred nits in her lifetime
- A single female head louse produces between eighty and one-hundred.
- Blood is sucked by the pumping action of pharynx.
- Adults as well as young ones of both the sexes feed on blood.
- The mouthparts are arranged in the form of fine stylets which, except when feeding, are withdrawn within the head.
- Blood is obtained by means of the stabber which is thrust out and then slowly pushed beyond the flesh.
- A salivary secretion is also injected which prevents the coagulation of the host's blood
- This helps the insect to suck in blood without forming the clots.
- The adults can feed on up to one milligram of blood at a time.
- The adults cannot survive for long without host. Hence, they prefer to take smaller quantities at frequent intervals.
- However, head louse can survive starvation for approximately three days at 30°C and five days at 24°C.
- On taking a very large meal, blood oozed out from the anus.
- Sometimes, overfeeding leads to gut rupturing.
- Adults are capable of fairly rapid active movement.
- They have been recorded as moving at a rate of 9 inches per minute; but they never move in straight lines.

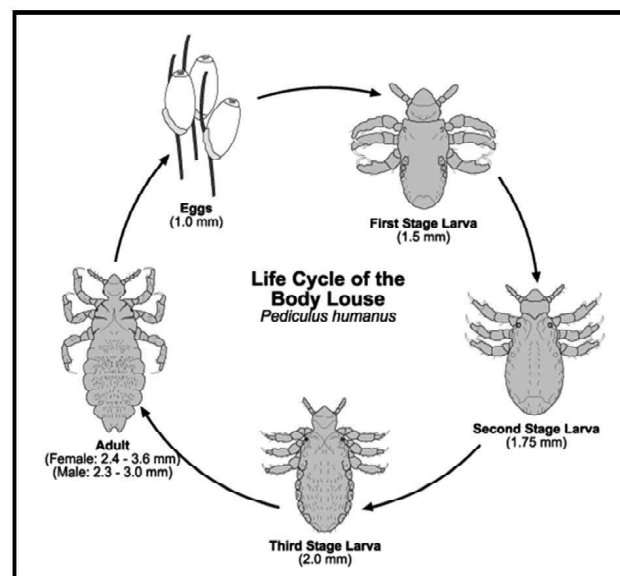


Fig. 4.7 Life Cycle of *Pediculus Humanus*

Figure 4.7 is depicting the generalized life cycle of *Pediculus humanus*.

Head louse is known to cause the following diseases:

1. **Epidemic Typhus:** The reservoir host of this disease are sheep and goats. Epidemic typhus is caused by a PPLO, *Rickettsia prowazeki*, which proliferates/increases in the gut of lice. Spores are released via faeces within 5 days of infection and can remain viable for up to four months in the dry conditions. *Rickettsia prowazeki* enters into the human system via contact with blood, wounds, conjunctive or by inhalation into lungs. The disease fatality rate is almost 100%.
2. **Trench Fever:** Trench fever is caused by *Rickettsia quintana*. This disease was prominent among soldiers during the World War II. Spores are released via faeces within 5 days of infection and can remain viable for up to four months in the dry conditions. *Rickettsia quintana* enters into the human system via contact with blood, wounds, conjunctive or by inhalation into lungs.
3. **Relapsing Fever:** Relapsing fever is transmitted via spirochaeta, *Borrelia recurrentis*. This breeds in the haemolymph of the louse and escapes when the louse is crushed or dies due to the parasitic infection. It gets into the human blood via wounds or scratches. This disease was prominent during the World War I and II.
4. **Vagabond Disease:** The common symptoms of this disease are itching, rashes and discoloration of the skin. Such symptoms occurs due to the allergic reaction to the bites and blood sucking by lice.

Control of head louse can be done as follows:

- Cleanliness and maintaining personal hygiene eradicate lice.
- Washing the hairs with anti-lice shampoo.
- Application of kerosene mixed with olive oil in equal ratio on the head kills lice.
- Lindane ointment, any insecticide mixed with oil kills all lice on head.
- The application has to be repeated every week to kill nymphs emerging from eggs.
- Body lice can be controlled by steaming or boiling of clothes and bedding or rinsing clothes in weak insecticide solution.

(b) The Crab louse (*Phthirus pubis*) as ectoparasite

- Crab louse (*Phthirus pubis*) is limited to the pubic region of human beings.
- Thoracic region of crab louse is very broad and abdomen is narrower when compared to the thorax
- Crab louse is five-segmented
- It bears small paired appendages.
- Crab louse are very sluggish lice
- Fecundity is approximately 50 eggs/female.

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- The eggs hatch in a time span of 7-8 days
- Nymphs generally take 27-33 days to develop into adults.
- Nymphs are very similar to the human louse.
- Body of crab louse is much broader when compared to its length.
- Legs appear to attach to the edge of the somewhat flattened body rather than to the underside.
- Crab louse moves very slowly
- Crab louse are named so due to their crab like motion
- As the name suggested, it infests hairs of pubic region as well pre-anal region of both sexes.

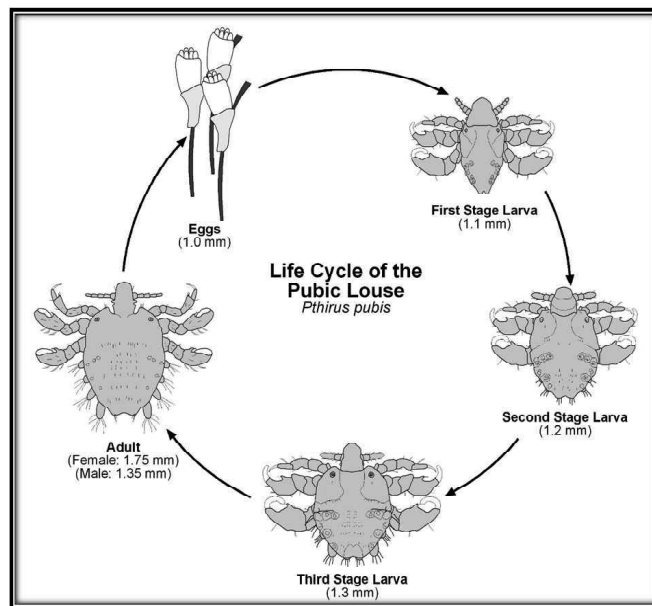


Fig 4.8 Life Cycle of *Pthirus Pubis*

Figure 4.8 is depicting the life cycle of *Pthirus pubis*

- Current worldwide prevalence of crab louse is estimated to be around 2% of the human population, however, accurate numbers are difficult to report as crab louse infestations are not considered a serious medical condition by health authorities. Most of the cases can be easily self-managed.
- Maintenance of hygiene has played a crucial role in reducing the worldwide population of crab louse.
- They are essentially found on the pubic region, however, infestation can also be seen on coarse hair elsewhere on the body (for instance: eyebrows, eyelashes, beard, moustache, chest, armpits, etc.).
- Crab louse do not generally occur on the finer hair of the scalp.
- Crab lice attach to pubic hair that is thicker than other body hair as their claws are adapted to the specific diameter of pubic hair and other thick hairs of the body.

- Crab louse infestations (phtiriasis) generally occurs via sexual contact and are most common in adults.
- The crab louse can travel up to 25 cm (10 in) on the body.
- Crab louse infestation is found worldwide and occurs in all races and ethnic groups and in all socio-economic levels.
- They can also be transmitted by close personal contact or contact with articles such as clothing, bed linen, and towels that have been used by an infected person.
- Crab louse present on the head or eyelashes of children might indicate sexual exposure or abuse.
- The main symptom of infestation with crab lice is itching, usually in the pubic-hair area, resulting from hypersensitivity to louse saliva, which can become stronger over two or more weeks following initial infestation.
- In some infestations, a characteristic grey-blue or slate coloration appears (*maculae caeruleae*) at the feeding site, which may last for several days.
- Crab lice are not known to transmit disease; however, secondary bacterial infection can occur from scratching of the skin.
- Crab louse infestation can be diagnosed by identifying the presence of active stages of the louse, as well as of eggs (nits) on the pubic hair and other hairs of the body.
- When infestation is diagnosed, other family members and contact persons should also be examined.
- A magnifying glass or dermoscope could be used for better identification.

To control crab louse following measures can be adopted: -

- All sexual contacts of the infected person should be examined carefully.
- All those who are infested should be treated.
- Sexual contact between the infested person(s) and their sexual partner(s) should be avoided until all have been examined, treated as necessary, and reevaluated to rule out persistent infestation.
- Washing all the clothes used by the infected person with hot water at least 130°F.
- Do not share clothing, bedding, and towels used by an infected person.
- Avoid using fumigant sprays or fogs as they can be toxic if inhaled through skin.
- Persons with pubic lice should be examined and treated for any other sexually transmitted diseases (STDs) that may be present.

(c) Bed-Bugs as Ectoparasite (*Cimex lectularis*; *Cimex hemipterus*)

- *Cimex lectularis* is prevalent in temperate countries while *Cimex hemipterus* (= *C. rotundatus*) is found in tropical countries.
- They are not known to transmit any major disease; however, they are supposed to be vectors for disease like kala-azar and relapsing fever.

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- The body of the bed bug is reddish-brown in colour and dorsoventrally flattened.
- The proboscis region of the bed bug is segmented and is used for piercing the host skin as well as for sucking blood.
- The maxillary palps are absent.
- Antenna of the bed bug is four-segmented.
- Pronotum is shield like having lobes on either side of the head.
- Tarsis is three-segmented.
- Wings are absent; however, rudimentary pads can be seen.
- There are present stink glands all over the body
- Stink glands emit a characteristic foul odor.
- In male, posterior end of abdomen is produced into a lobe like structure and is hairy with the presence of a spine-like aedeagus.
- The abdominal region of the female is broadly rounded posteriorly having small hairs.
- Fourth abdominal segment in female bears a specialized structure known as organ of Berlese on the ventral side to the right side of the middle.
- Organ of Berlese is a pouch-like structure in which sperms are stored after copulation.
- Sperms from here bore via the walls as well as tissues and fertilize eggs in the ovaries.
- Eggs are whitish in colour, 1.0 mm long, flask-shaped structure, having reticulated surface and an operculum on the anterior side.
- Bed bugs are placed among the bedding, in cracks or crevices of the walls as well as floor or they can be seen in any other hidden places.
- Many females lay their eggs in groups at one place.
- Fecundity of the female is approximately 75-200 eggs/female.
- The incubation period varies from six to 10 days, however, in hot weather conditions it get reduced.
- Nymphs are very much similar to the adults and suck blood.
- First instar nymphs are semi-transparent whereas later instars are straw yellow in colour.
- There are five nymphal instars which take approximately thirty days to become adults.
- Adults are nocturnal
- Both the sexes suck blood.
- They can tolerate starvation up to a period of six months.
- The starving individuals look light in colour and are extremely thin.
- They can regain dark brown in colour and original form after sucking the blood.

Note: Bed bugs take 3-10 minutes to complete feeding

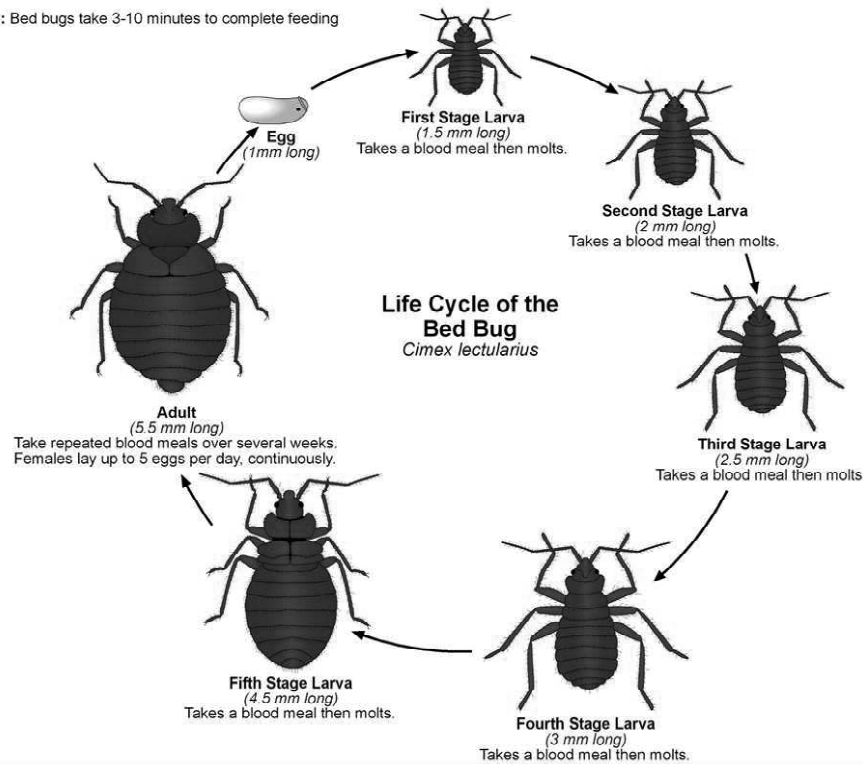


Fig. 4.9 Life Cycle of Bed Bugs

Figure 4.9 is depicting the life cycle of bed bugs.

Control of Bedbugs

Regular maintenance of cracks and cervices on the floor as well as walls.

- Regular white washing of the house.
- Maintenance of cleanliness also helps to get rid of bugs.
- Periodically exposing the bedding as well as other infested material to the sun.
- Washing the clothes with warm water.
- Spray of DDT 5%, BHC 6% or Malathion and Endosulfan dust 5% in houses and cots and beds kills them.
- Dusting helps to eradicate bed bugs more effectively.

Predators of Bed Bugs

Small red house ant, *Monomorium pharaonic*; *Rduvius personatus*, the assassin bug; The pseudoscorpion, *Chelifera cancrroids*.

(d) Fleas as Ectoparasite

- Fleas are small wingless insects.
- The length of the flea is approximately 2-3 mm long.
- The fleas are reddish-brown in colour.
- The fleas are highly sclerotised, having laterally compressed bodies.

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- Antennae of the fleas are short, stout, pectinate or clubbed and concealed in a groove.
- Mouth parts of the fleas are modified for piercing and sucking action.
- Legs of the fleas are adapted for clinging with curved claws.
- Hind leg is longer and modified for jumping.
- Body of the flea's bears backwardly directed spines that help them to move through the hairs of the hosts.
- The abdomen of the male is directed upwards at the apex and flat.
- Apical part of the abdominal region bears a coiled cirrus organ which serve as the copulatory organ of the male.
- The abdomen of the female is directed straight backwards and carries a small pouch-like structure known as spermatheca
- Spermatheca bears sperms after copulating with the male.
- Fleas breed in dark and secluded places having high moisture contents like dusty floors, under carpets, rat burrows, granaries, chicken houses as well as godowns.
- Female lays approximately 300-400 eggs in its lifetime
- The incubation period varies from 2-3 days (maximum 12 days).
- Larvae is yellowish-brown in colour, caterpillar-like, 13-segmented, hairy
- Larvae move actively, feeding on dead and decaying organic matter as well as on faeces of the adult fleas.
- Larva is approximately 4 mm in length and lack eyes, antennae as well as legs
- Larvae bears long bristles on the body, which also help the fleas in locomotion.
- There are 3 instars and larval period ranges between 7 to 10 days.
- Pupation of the fleas takes place in a very tough, dull whitish cocoon that is concealed in dust particles.
- Pupal period is approximately seven days.
- Pupa may hibernate for up to a period of one year.
- The entire life cycle of the fleas is completed in about 20 days under favourable conditions.
- After emergence, adults hop on to the passing rats or other hosts and start sucking blood.
- Rats usually sucks in more blood than necessary
- A lot of undigested blood passes along with faeces that form food for the larvae.
- An adult can jump up to 8 inches high and 13 inches in length.
- Longevity of the adult is approximately two years
- It can tolerate/resist starvation up to a period of six months.

There are about 200 species of fleas in India. A few well known common species have been explained below:

- 1. Human Flea (*Pulex irritans*).** Human flea is a cosmopolitan species, which is commonly found in hilly areas. Man is the preferred host, however, it can feed on other animals as well. Human flea is a combless flea. This species of flea is not a vector of plague. Its mesopleuron is without a ridge like crest or carina.

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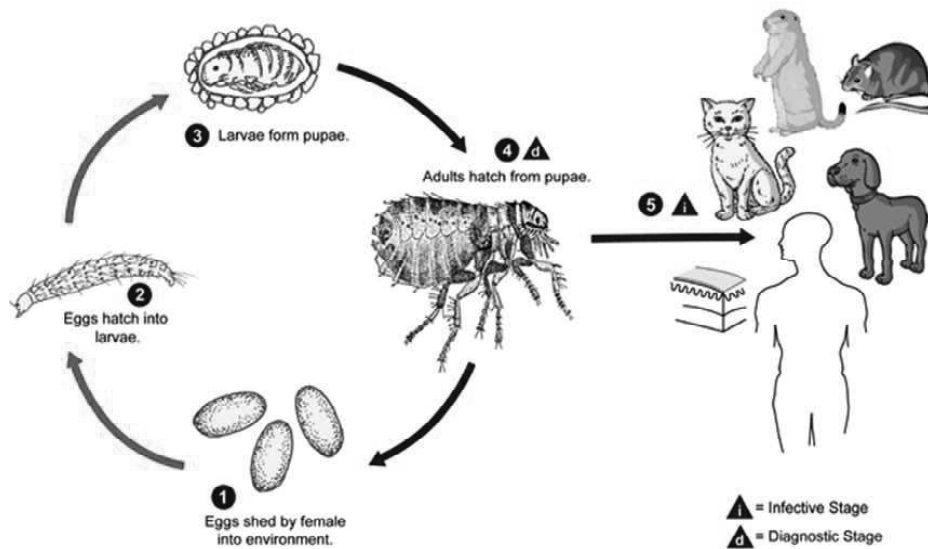


Fig. 4.10 Life Cycle of Flea

Figure depicting the life cycle of flea

- 2. Rat Flea (*Xenopsylla cheopis*).** Rat flea is a combless flea seen on rats and act as the vector of dreaded plague. It resembles human flea; however, mesopleuron has a crest or carina located above the middle coxa. Rat flea breeds in rat dwellings and repeatedly bites human beings.
- 3. Dog and Cat Flea (*Ctenocephalides canis* and *C. felis*).** Unlike human and rat fleas, dog and cat fleas are combed fleas present on dogs and cats. They bear both the genal and pronotal combs and spines all over the body.
- 4. Combed Rat Flea (*Nosopsylla* or *Ceratophyllus* sp.).** Combed rat flea is similar to the other rat fleas however it bears a pronotal comb. Genal comb is absent. The European species *N. fasciatus* is a vector of plague however the Indian species, *N. nilgiriensis* is not a vector of plague.
- 5. Broken-Headed Mouse Flea (*Leptopsylla segnis*).** Broken headed louse is present on mice. Both genal as well as pronotal combs are present. Eyes are absent. The antennal grooves of both sides join on the dorsal side, giving a broken head appearance or it seems as the head is divided into two parts.
- 6. Rat flea (*Stivalius ahale*).** Rat flea is prevalent in the foot hill of south India. It resembles *Nosopsylla*, however, it is larger having round head, lot of bristles and reduced eyes. It has only pronotal comb and is a vector of plague in south India.

Disease Transmission

Fleas majorly transmit plague that is caused by the safety pin bacillus called *Yersinia pestis* or *Pasturella pestis*. Plague is caused in man and in rats equally and produces three types of symptoms in man.

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- 1. Bubonic plague:** The bacillus infects lymphatic system, leading to swelling and pain in the lymph glands, however, no fever occurs. Rats also suffer from similar kind of symptoms.
- 2. Septicemic plague:** Septicemic plague spreads to the blood vascular system causing fever. It causes headache as well as pain in the back. Sudden chilliness, redness of eyes, rapid pulse, thick speech and high fever are other symptoms. In the case of prolonged illness, spleen enlarges in size and becomes brick-red in colour and liver is also enlarged. Coma and death are quite possible.
- 3. Pneumonic plague:** Pneumonic plague multiplies in the lungs and pleural cavity causing pneumonia-like symptoms. A Yellowish colour fluid fills the lungs and pleural cavity, leading to excessive coughing as well as heavy breathing. Infection can spread directly from man to man via droplets which are released during coughing. Pneumonic plague is one of the most dangerous kind of plague as it spreads rapidly by droplet infection, chiefly in areas having high population density and leads to quick deaths.

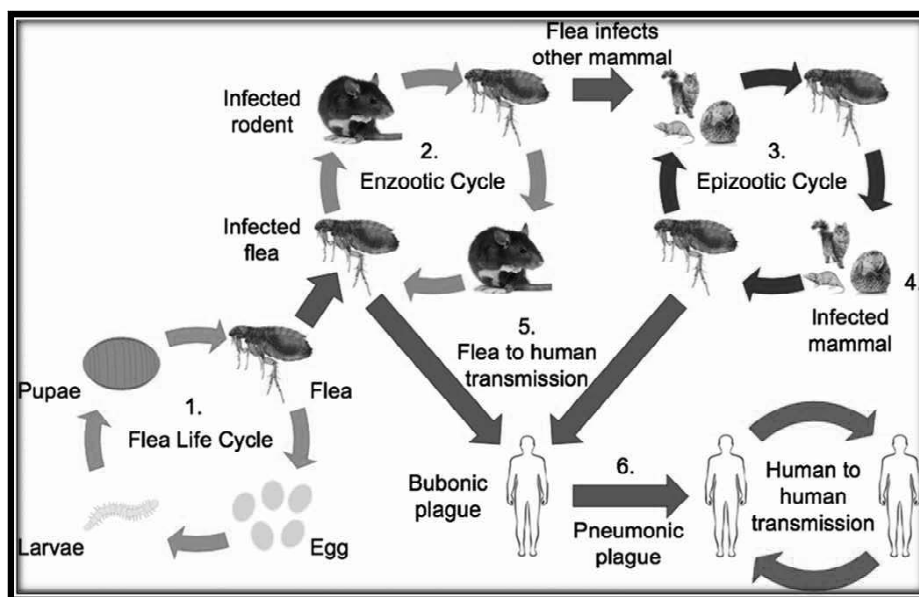


Fig.4.11 Transmission of Disease by Fleas.

Figure 4.11 is depicting the transmission of disease by fleas.

Mode of infection

- Whenever, a flea sucks in blood from a rat infected with plague, it gets bacillus into the crop region along with the blood meal.
- The bacilli get attached to the spines of proventriculus and form mucilaginous colonies there, multiplying very fast, blocking the passage of blood into the intestine.

- Such fleas are referred to as *blocked fleas* that always remain hungry as the blood does not pass into the intestine.
- Hence, they keep biting and sucking blood constantly but regurgitating it in the same wound as the stomach is already full of blood and bacilli.
- In doing so, they keep transmitting bacilli from their stomach to the new hosts.
- Blocked fleas can survive up to 40 days and transmit disease continuously.

Murine Typhus

- Murine typhus is another disease transmitted by *Xenopsylla cheopis*.
- It is a mild typhoid fever caused by the PPLO, *Rickettsia mooseri*.
- The organism responsible for causing murine typhus multiplies in the gut of flea and is excreted out via faeces.
- Man gets infected either by contamination of wounds or by the flea faeces or by inhalation of dust containing faeces.
- A few fleas act as intermediate hosts for the tape worms like, *Dipylidium caninum* and *Hymenolepis diminuta*.

Control of Fleas

- Controlling rats is an effective method of controlling fleas.
- The effective methods of controlling rats include Trapping, baiting and fumigation.
- Fumigation by cyanogas kills not only rats but also all stages of fleas in the rat burrows.
- Fumigation should be done regularly after two three months.
- Construction of rat proof godown having metalled doors as well as meshed windows is also an effective method to keep the rats away from human dwellings.
- Dusting the houses, floors, godowns as well as other places visited by rats must be done on regular basis using residual insecticides like BHC, endosulfan, dieldrin, aldrin etc.
- This will kill all stages of fleas as they breed in dust and abandoned corners.
- Patients suffering from plague can be treated with streptomycin injections or oral doses of antibiotics such as tetracycline, sulphadiazene, chloramphenicol, doxycycline, azithromycin etc. given two or three times in a day.

(3) Insect as Vector of Human Diseases:

(a) Mosquito as vector of human diseases

- Mosquitoes are small approximately 15 mm in length
- They are two-winged insects.
- Mosquitoes can be differentiated from other flies due to the presence of following characteristics: a long proboscis projecting in forward direction

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- from the head; the presence of scales on the wing veins; presence of fringe of scales along the posterior margin of the wing; and a characteristic wing venation, the second, fourth and fifth longitudinal veins being branched (Goma, 1966).
- Males and females can be differentiated by the different structure of the antennae.
 - In males, plumose like antennae are present
 - In females, antennae bears only a few short hairs.
 - Genera and species identification can be performed by standard entomological aspects such as exterior, arrangement of eggs, habitus etc.
 - Further and newer techniques include biochemical techniques such as enzyme electrophoresis and gas chromatography of cuticular carbons, and DNA probes.
 - Mosquitoes are holometabolous insects showing all the four different stages i.e., egg, larvae, pupa and adult.
 - Most of the common and important mosquitoes act as transmitters of pathogens for animal as well as man belong to four different genera namely: *Anopheles*, *Culex*, *Mansonia* and *Aedes*.
 - Mosquitoes bears sensory organs in their antennae to recognise host and oviposition sites.
 - The Johnston's organ in the basic segment by which males recognise wing beats of the females.

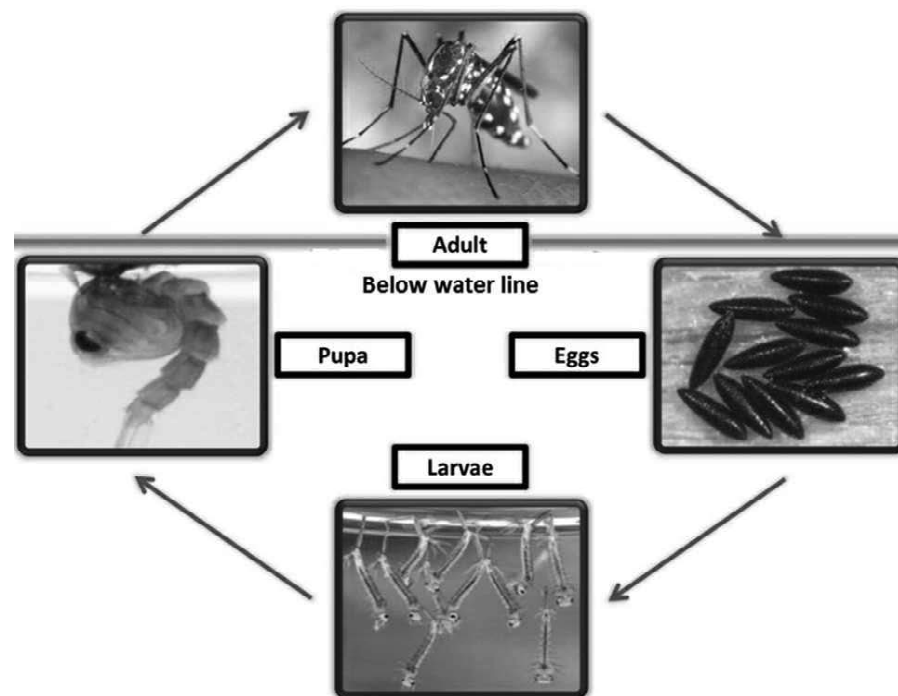


Fig 4.12 A Generalized Life Cycle of Mosquito Showing Different Stages i.e., Egg, Larva, Pupa and Adult

Figure 4.12 shows a generalized life cycle of mosquito showing different stages i.e., egg, larva, pupa and adult.

Identification Characters of Mosquito

- Males have rudimentary maxillae and mandibles; hence they fail to suck blood but can easily suck fluids and nectar from flowers.
- Males also possess very bushy whorl plumose antennae and tip of abdomen with typical male genitalia.
- Females have short hairs on the antennae and needle-like maxillae and mandibles for piercing the skin of host for sucking blood.
- Other characteristics are summarized below according to the species.

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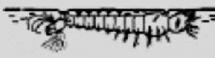
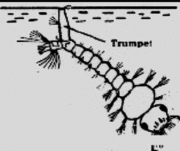




| SUBFAMILIES | | | |
|---|--|---|--|
| Anopheline | Culicine | | |
| Genus Anopheles | Genus Aedes | Genus Culex | Genus Mansonia |
| Eggs | | | |
|  float |  do not float |  raft of 25 - 100 eggs |  Aquatic plant |
| Larval stage | | | |
|  | |  Trumpet | |
| Pupal stage | | | |
|  |  | | |
| Adult stage | | | |
|  Long palps |  Short palps | | |

Fig. 4.13 Different Stages of Life Cycle of Mosquito

Figure 4.13 depicting characters of different stages of life cycle of mosquito.

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Types of Malaria

- **Tertian, Benign Tertian or Vivax Malaria:** *P. vivax* is the causative agent of this type of malaria. The characteristic feature of this type of malaria is the recurrence of fever every third day i.e. 48 hrs. It is mainly confined to temperate regions
- **Quartan malaria:** The causative agent of this type of malaria is *P. malariae*. The characteristic feature of this type of malaria is the recurrence of fever every fourth day i.e. 72 hrs. It is mainly confined to tropical and subtropical regions.
- **Ovale or Mild Tertian Malaria:** The causative agent of this type of malaria is *P. ovale*. The characteristic feature of this type of malaria is recurrence of malaria every 48 hrs. It is mainly confined to tropical Africa.
- **Aestivo-Autumnal, Malignant Tertian or Pernicious Malaria:** This type of malaria is caused by *P. falciparum*. The fever cycle is of 48 hrs. The fever is often fatal to patient as it affects the brain.
- **Quotidian Malaria:** This type of malaria occurs when the patient is affected by more than one species of plasmodium or when 2 or 3 generations of parasite mature on successive days. The fever is repeated daily in this infection.

Symptoms and Pathogenesis

In malaria, attack of fever shows three successive stages:

- **Cold stage:** In this stage, the patient suffers from bone chilling cold. It lasts for 20 mins to 1 hr.
- **Hot stage:** As the chill subsides, the temperature of the body rises to 41°C. The patient feels hot with headache. It lasts for one to four hours.
- **Sweating stage:** As the temperature declines, the patient sweats profusely. Due to this, fever comes down until the next cycle of fever.

Malarial fever occurs when schizonts in RBCs burst and liberate contained merozoites and malarial pigment, i.e., haemozoin in blood plasma.

Anaemia: Anaemia occurs in malarial fever due to following reasons:

- (a) Destruction of RBCs on release of merozoites
- (b) Infected RBCs becomes fragile
- (c) Malarial parasite produce haemolysin which cause haemolysis of the RBCs
- (d) Spleen affected by malarial infection releases lysolecithin which destroys RBCs

Control of Malaria

All the control measures for malaria fall into three categories

(1) Elimination of Vector i.e. *Anopheles* Mosquito

(a) Anti-larval Measures

- Removal of breeding places for mosquito such as pots, old tyres, tins and other containers

- Covering of water surface with oil, petroleum, kerosene oil, crude oil or paraffin oil.
- Water storage tanks, coolers should be cleaned and treated periodically
- Do not allow water to be stagnant
- Biological control of larvae and pupae in ponds has been achieved by releasing larvivorous fishes and insects, such as the native *Gambusia* and adult insects like dragon-flies. These fishes and adult insect actively feeds on the larvae of mosquito

(b) Anti-Adult Measures

- Spraying insecticide
- UV electrocuting traps should be used to attract and kill adults.
- Destruction of tall grasses and bushes which serves as local breeding ground for adults.
- Aerial sprays of pyrethrum, carbaryl, carbofuran, arprocarb mixed with mineral oil are still effective in killing adults.

(2) Prophylaxis, i.e., Prevention of Infection

- Use of effective mosquito repellent such as citronella oil, dimethylphthalate, odomos cream or pyrethrum cream.
- Use of mosquito nets.
- Frequent use of mosquito repelling fumigants.
- Screening of doors, windows and good ventilation at home'.
- Taking proper nutrition and avoiding contact with bad conditions.

(3) Treatment of the Patient

- Treatment of *P. vivax*: Drugs like Primaquine may be given under medical supervision.
- Treatment of *P. falciparum*: Drugs like Chloroquine may be given under medical supervision. However, if the patient has developed resistance to the above mentioned drug ACTs are recommended. The ACT (artemisinin-based combination therapies). The ACT used in India is artesunate + sulfadoxine-pyrimethamine.
- Treatment of Complicated Malaria: Administration of intravenous or intramuscular injection for a minimum of 24 hrs.

Mosquito Control

It is difficult to achieve successful mosquito control measures as they develop resistance against insecticides very quickly. Moreover, they are able to inhabit a variety of environmental conditions. Still, some measures have been adopted to reduce mosquito populations.

(A) Personal Preventive Measures

- Use of effective mosquito repellent such as citronella oil, dimethylphthalate, odomos cream or pyrethrum cream

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- Use of mosquito nets
- Frequent use of mosquito repelling fumigants

(B) Anti-larval Measures

- Removal of breeding places for mosquito such as pots, old tyres, tins and other containers.
- Water storage tanks, coolers should be cleaned and treated periodically.
- Do not allow water to be stagnant.
- Biological control of larvae and pupae in ponds has been achieved by releasing larvivorous fishes, such as the native *Gambusia*. *These fish actively feeds on the larvae of mosquito*

(C) Anti-adult Measures

- Spraying insecticide.
- UV electrocuting traps should be used to attract and kill adults.
- Destruction of tall grasses and bushes which serves as local breeding ground for adults.
- Aerial sprays of pyrethrum, carbaryl, carbofuran, arprocarb mixed with mineral oil are still effective in killing adults.

There are more than 3000 species of mosquitoes in the world which are virtually distributed everywhere except in Iceland and poles. The table 4.5 below lists a few mosquito species which are responsible for transmitting deadly human diseases:

Table 4.5 Mosquito Species

| Genus | Breeding site | Place found | Disease and distribution |
|---|--|------------------|--|
| <p>Anopheline mosquitoes</p> <p>Anophelines breed in non polluted water</p> <p>Biting period : NIGHT</p> | <p>edges of rivers, swamps, impoundments, ditches, tanks, saltwater habitats protected from wave action, rice fields, temporary rainpools, hoofprints.</p> | <p>Worldwide</p> | <ul style="list-style-type: none"> ■ Malaria: Tropical and sub-tropical areas ■ Bancroftian filariasis: Asia and Africa ■ Brugian filariasis: Asia ■ O'nyong nyong virus: Africa |
| <p>Aedes mosquitoes</p> <p>One species lives in close association with man, in any kind of human settlement. The <i>A. aegypti</i> breeds in any small water collection.</p> <p>Aedes spp. are primarily forest mosquitoes.</p> <p>Biting period : DAY</p> | <p>Tin cans, plastics, car tyres, gutters, ornamental ponds, tanks, jars, any type of container, waste disposal areas, tree holes.</p> | <p>Worldwide</p> | <ul style="list-style-type: none"> ■ Yellow fever: Africa and Americas ■ Dengue: Africa, Americas, Asia ■ Dengue Haemorrhagic fever: Americas, Asia ■ Bancroftian filariasis: Pacific ■ Other arbovirus: Africa, Americas, Asia |

| | | | |
|---|---|--|---|
| <p>Culex mosquitoes</p> <p><i>C. quinquefasciatus</i> breed in any dirty water in urban and rural areas. Other species are also very common in rice fields in Asia.</p> <p>Biting period : NIGHT</p> | <p><i>C. quinquefasciatus</i></p> <p>Waste water ditches, latrines, septic pits, cesspools, drains, waste disposal.</p> | <p>Worldwide</p> | <ul style="list-style-type: none"> ■ Bancroftian filariasis: Most tropical areas ■ Encephalitis virus: Africa, Americas, Asia, Europe |
| <p>Mansonia mosquitoes</p> <p>Mainly associated with aquatic plants, in rural areas where irrigation canals occur.</p> <p>Biting period : NIGHT</p> <p>It is a vicious biter</p> | <p>Ditches, ponds, irrigation canals, swamps.</p> | <p>Essentially tropical</p> <p>Worldwide</p> | <ul style="list-style-type: none"> ■ Brugian filariasis: Asia ■ Other arbovirus: Rare in Africa and Americas |

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(b) Sand Flies as Vector of Human Disease

- Sand flies are small insects.
- The length of the sandflies is approximately 1.2-3.7 mm (Perfiliev, 1968).
- Head, thorax, abdomen and appendages are covered with hairs which give a “fluffy” appearance to the sand flies.
- The palps reach beyond the proboscis region and are nearly folded in two.
- The wings are raised above the body.
- The apex of the wings is directed posterior and lateral.
- Legs are long and slender in structure (Perfiliev, 1968).
- Sand flies which are disturbed on a wall do not usually fly, however, they jump away, rising to the ceiling.
- This jumping behaviour, the raised wings and the “fluffiness” are reported to be so characteristic that recognition of a live sand fly and distinguishing it from other small insects is possible (Perfiliev, 1968).
- Females and males of the sand flies are easily distinguishable.
- In females, the end of the abdomen appears blunt.
- The abdomen is narrow immediately after hatching, however, it gets inflated and reddish in colour after the first blood meal.
- The color of the abdomen changes brown to black after digestion of the blood and it becomes characteristically thicker with a yellowish shade during egg development.
- The male abdomen resembles that of unfed females or even thinner and ends in paired appendages, appearing like small spines to the naked eye (Perfiliev, 1968).
- Besides anatomical structures, techniques like isoenzyme electrophoresis, gas chromatography, monoclonal antibodies and DNA probes are used for species identification and differentiation.

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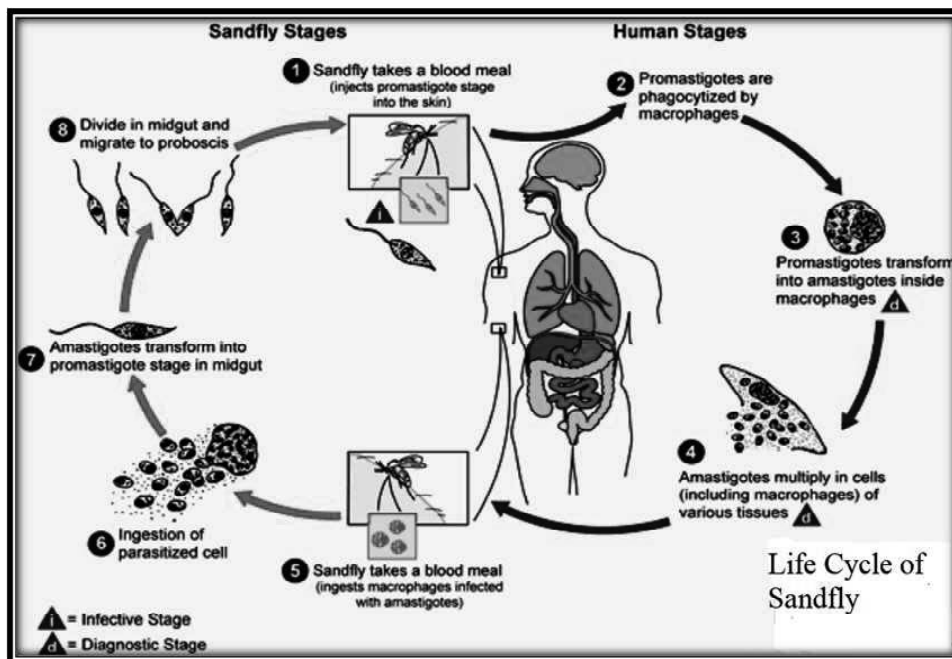


Fig. 4.14 The Life Cycle of Sandfly

Figure 4.14 is depicting the life cycle of sand-fly.

There are about 33 species of sand flies in India, some of which are known to transmit diseases. The table 4.6 below lists some sand fly species which are known to transmit deadly diseases.

Table 4.6 Some Sand Fly Species and Diseases Transmitted By Them

| Sandfly species | Disease name | Causative organism | Distribution |
|---|---|---------------------------------|--------------|
| <i>Phlebotomus argentipes</i> | Kala-azar, | <i>Leishmania donovani</i> | NE India, TN |
| <i>P. sergenti</i> ; <i>P. papatasi</i> | Oriental Sore | <i>Leishmania tropica</i> | C & N India |
| <i>Phlebotomus papatasi</i> | Sandfly fever | Virus | Whole India |
| <i>P. intermedius</i> | Espundia (naso-Pharyngeal leish-maniasis) | <i>Leishmania braziliensis</i> | S. America |
| <i>P. varrucarum</i> | Carrion's disease | <i>Bartonella bacilliformis</i> | S. America |

Sand Fly Control

- Spraying the houses with malathion, endosulfan, carbaryl etc. kills the sand fly adults and other stages.
- Maintaining cleanliness in houses and surroundings and
- Plastering of cracks and crevices also reduces adult population.
- Spraying of breeding places with insecticides is an effective control measure.
- It is difficult to control larval population as they burrow deep into the soil, however, treating the garbage dumps with insecticides helps in controlling the population.

(c) Housefly (*Musca domestica* and *Musca vicina*)

House fly is a larger insect, approximately 6-7 mm long, greyish in colour with blackish markings on the body. It has two wings with a wing span of 13-15 mm and posterior wings being modified in a pair of balancers or halteres.

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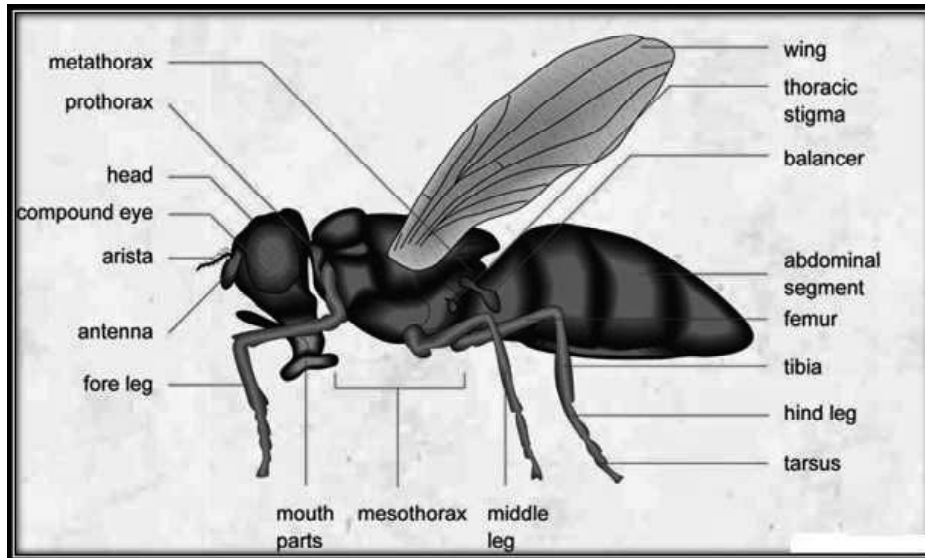


Fig. 4.15 Morphological Structure of Housefly

Figure 4.15 depicting the morphological structure of housefly.

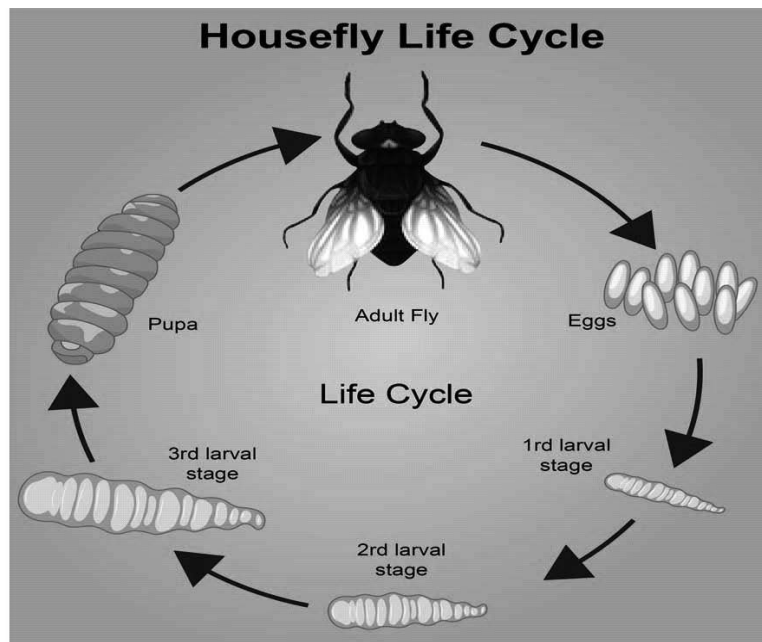


Fig. 4.16 Life Cycle of Housefly

Figure 4.16 is depicting the life cycle of housefly.

The table 4.7 lists the disease transmitted by house fly.

Table 4.7 Disease Transmitted By House Fly

| Disease | Causative organism |
|--|---------------------------------|
| 1. Cholera | 1) <i>Vibrio cholerae</i> |
| 2. Typhoid | 2) <i>Salmonella typhi</i> |
| 3. Paratyphoid | 3) <i>Salmonella paratyphi</i> |
| 4. Amoebic dysentery | 4) <i>Entamoeba histolytica</i> |
| 5. Bacillary dysentery | 5) <i>Shigella dysenteriae</i> |
| 6. Poliomyelitis | 6) <i>Polio virus</i> |
| 7. Trachoma eye disease | 7) <i>Virus</i> |
| 8. Eggs of many species of Parasitic worms | 8) Cestodes and nematodes |

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Mode of Transmission of Diseases

- **External carriage:** Pathogenic organism attaches to the lower side of oral disc, abdomen and hairs of tibia of housefly. These pathogenic organisms are carried to the food and water by housefly and finally enter into human system through contamination.
- **Internal carriage:** In this mode of transmission, pathogens are swallowed and retained in the gut. As the gut holds the pathogens, housefly can transmit these pathogens either by defecating or regurgitating on the food.

Control of House Flies

- Removal of breeding places: Maintenance of cleanliness is an effective method to bring down population of house fly.
- Treatment of breeding places: Cover the breeding grounds with a layer of insecticide dust such as 5% DDT to prevent flies to feed and lay eggs.
- Adults can be killed by spraying insecticide such as malathion, Baygon, , lindane, carbaryl or endosulfan
- UV emitting electrocuting trap are quite effective in attracting and killing adults
- Biological control: *Spalangia*, has been found to be very effective in destroying pupae of houseflies.

(d) Tsetse Fly

- Tsetse fly belongs to genus *Glossina* of family Glossinidae and is pronounced **tse-tse** or **teet-see** or **set-see**.
- Tsetse are large biting flies which are found in Africa
- Tsetse flies suck blood of vertebrates and transmit numerous protozoan diseases.
- Tsetse flies can be differentiated from other flies as they fold their wings completely while resting in such a way that one wing rests directly on top of the other.
- The color of the adults varies from yellowish to brownish in colour.
- Adult possess a piercing proboscis as well as a hatchet-shaped cell in the centre of each wing.

- The arista of the third antennal segment has branched setae.
- Larvae of tsetse flies breathe via a pair of posterior spiracles.
- Third instar larvae respire via a pair of lateral lobes, which comprises of three air chambers and open to outside via numerous spiracles.
- Female tsetse flies are viviparous.
- Female tsetse flies retains a single egg that develops to the third larval stage before being deposited.
- The egg within the uterus hatches in a time span of 3–4 days.
- The egg hatches to form first instar larva that obtains nourishment from secretions of a pair of uterine glands from the mother.
- The third instar larvae are deposited in soil.
- Inside the soil, they form burrows and keep on feeding and pupate for 4–5 weeks.
- The young adult emerges from the pupa and both sexes suck blood.
- Males remains infertile for several days after emergence.
- On the contrary, females are ready to copulate two to three days after emergence.
- The first larval offspring is deposited about 9–12 days after the female emerges.
- Tsetse flies females live up to fourteen weeks.
- Males live for approximately six weeks.
- Tsetse flies comprises of around thirty-four species and sub-species that are placed in a single genus known as *Glossina*.
- A few species of tsetse flies transmit trypanosome species that cause trypanosomiasis or **sleeping sickness** in humans caused by *Trypanosoma brucei gambiense* and *T. brucei rhodesiense*.
- In animals, they transmit **nagana**, in cattle, horses, pigs and antelopes which is caused by *Trypanosoma brucei brucei*, *T. congolense* and *T. simiae* and **surra** in horses and pigs caused by *Trypanosoma suis*. Tsetse transmits trypanosomes in two ways:
 - **Mechanical transmission** of the disease involves direct transmission of trypanosomes taken from an infected host into an uninfected host. In this, tsetse fly feeds on an infected host and acquires trypanosomes in the blood meal and then, if within a short period it feeds on an uninfected host and regurgitates infected blood and saliva into the tissue of uninfected animal, it has transmitted the protozoans.
 - **Biological transmission of the disease** requires a period of incubation of the trypanosomes within the tsetse stomach where the protozoans reproduce via several generations to become infective stage. Tsetse infected by trypanosomes are thought to remain infected for the remainder of their lives.

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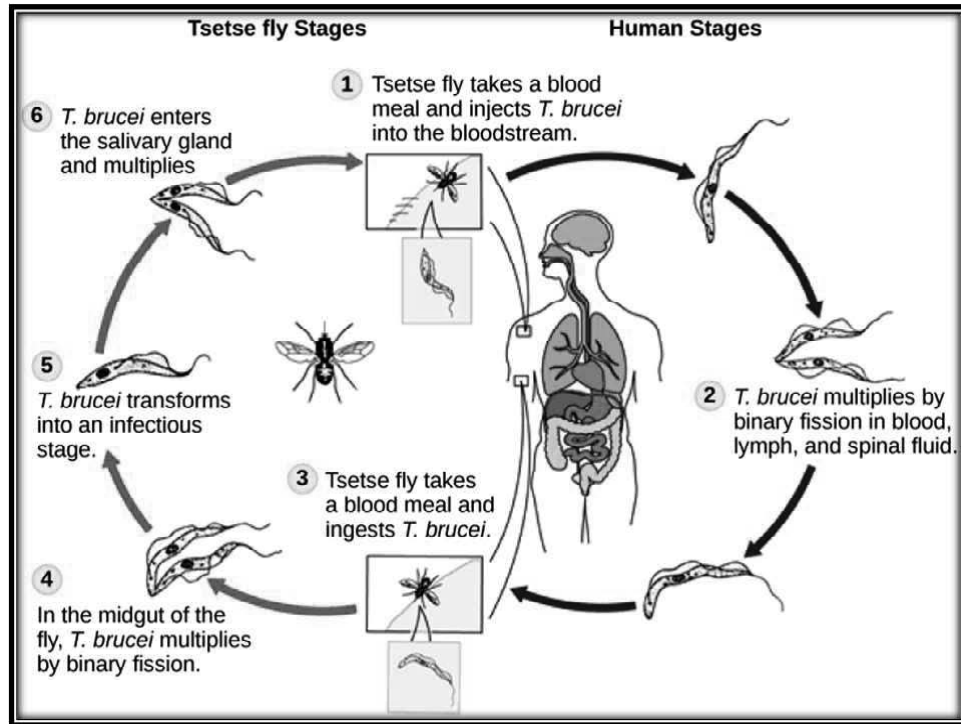


Fig. 4.17 Life Cycle of Tsetse Fly

Figure 4.17 is depicting the life cycle of Tsetse fly.

The table 4.8 lists the insect vectors along with their habit, habitat, distribution and the disease transmitted.

Table 4.8 Insect Vectors Along With Their Habit, Habitat, Distribution and the Disease Transmitted

| Vector (genus) | Particularity | Breeding sites and habits | Disease and distribution |
|---|--|---|--|
| Tabanid or Horsefly <i>1. Chrysops</i> | They are very robust Length; 6 to 10mm | Only the female feeds on any animal. She lays 100 to 1000 eggs according to species. They breed in moist and wet ground. | ■ <i>Loa loa</i> filariasis: West and Central Africa |
| Tsetse fly <i>Glossina</i> <i>Mortisans group</i> (savannah flies) <i>Palpalis group</i> (river bank flies) | Very long proboscis wide wings 9 to 25mm in length | Both males and females suck blood. Tsetse flies are viviparous. They deposit their larvae in damp ground and arid areas. | ■ Sleeping sickness: Africa |
| Sandfly <i>Phlebotominae</i> | Less than 3mm long Very long legs | The females only are blood-sucking at night. They are located in the tropics and subtropical areas south of Europe. They breed in moist and wet ground. | ■ Cutaneous and visceral (Kala Azar) Leishmaniasis occur in Sudan, Latin Americas, India, Asia, Middle East, and Southern Europe |

| | | | |
|---|---|---|---|
| <p>Bedbug</p> <p><i>Cimex spp.</i> Reduviid bugs</p> | <p>7mm long brownish insects</p> <p>flat and oval body</p> | <p>They lives in temperate and tropical zones. They are active only at night where they feed on humans and animals.</p> | <ul style="list-style-type: none"> ■ Bedbugs cause nuisance such as itchiness: Worldwide ■ Chagas disease is transmitted by triatomine bugs in South and Central Americas, and in some parts of Caribbean |
| <p>Blackfly</p> <p><i>Simuliidae</i></p> | <p>Small insect, 1 to 6mm in length</p> | <p>They bite during the day. Only the female sucks the blood of animals and humans. They breed and live in all kinds of unpolluted water, vectors in Africa breed only in fast flowing oxygenated streams or rivers. They have a worldwide distribution.</p> | <ul style="list-style-type: none"> ■ Onchocerciasis or river blindness: Africa, and some parts of Latin America |
| <p>Cockroach</p> | <p>5 to 73mm in length</p> <p>two pairs of wings</p> <p>flattened appearance</p> <p>Yellow-brown to dark colour</p> | <p>These insect are very agile and fast and live in colonies. They have a preference for man-made structures where it is warm. In the tropics they may live and breed outdoors. Latrines may be infested in a refugee camp. They are particularly active at night.</p> | <ul style="list-style-type: none"> ■ Cockroaches act as mechanical vectors and may transmit diarrhoeal diseases, typhoid fever, dysentery, viral diseases: Worldwide |
| <p>Tick</p> <p>Hard tick</p> <p>Soft tick</p> | <p>7 to 20mm in length</p> <p>Hard back</p> <p>Soft back</p> | <p>Both males and females feed on warm-blooded animals and humans. They are attracted by the carbon dioxide from their prey. Hard ticks are located in vegetation and soft ticks live in close association with available prey. They can survive several years of starvation.</p> | <ul style="list-style-type: none"> ■ Relapsing fever: Worldwide ■ Q-fever: Africa, Americas ■ Lyme disease Arbovirus diseases: Worldwide <p>Ticks are very painful biters and can cause serious loss of blood</p> |

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(3) Arachnids as Ectoparasite

(a) TICKS as Ectoparasite

A few ticks are as follows:

Boophilus annulatus

Ixodes ricinus

Hyalomma detritum

Haemaphysalis bispinosa

Amblyomma variegatum

- Ticks are usually whitish to bluish-grey in colour having an oval body structure wrapped inside a tough and wrinkled skin.

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- They possess four pairs of legs, a pair of chelicerae modified for piercing as well as sucking and a pair of pedipalps.
- Female tick separates from the host and lays eggs in soil
- The eggs hatch into a six-legged larval stage known as seed-tick.
- The seed tick grows in soil
- On a suitable chance/opportunity, it attaches on to the host and changes into a four-legged nymph which ultimately molts to become adult.
- It takes around 60 days to complete the life cycle
- Ticks are vectors of cattle fever, tick fever as well as several other pathogens.
- Infection by ticks leads to enlargement of spleen as well as discoloration of urine.
- Ticks prefer to suck blood from soft parts of the host body, like ear pinna or base of the tail.

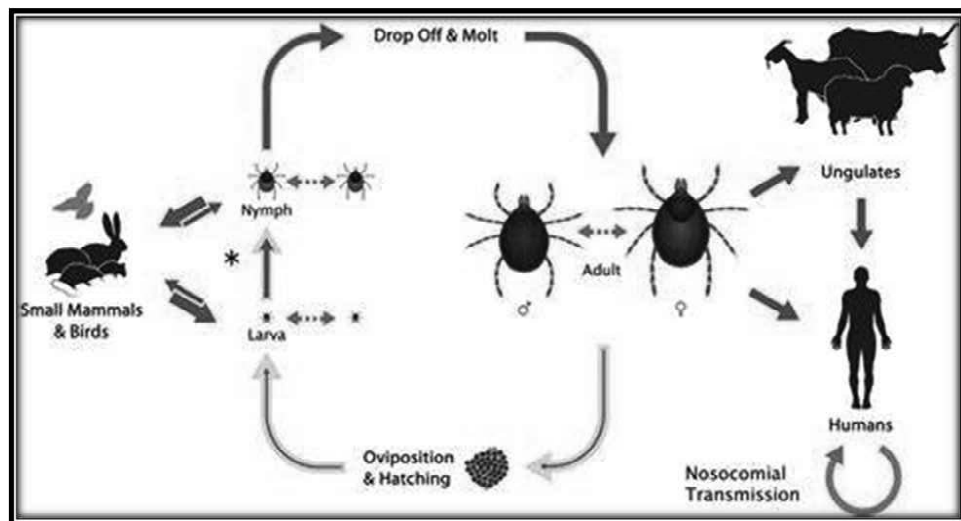


Fig.4.18 Life Cycle of Tick.

Figure 4.18 is depicting the life cycle of tick.

Diseases Transmitted

The following disease are transmitted by tick infection.

- **Bovine piroplasmosis** (caused by the protozoan, *Babesia* species).
- **Horse tick fever** is caused by 2 species of *Babesia*.
- **Theileriosis** is a disease of goat and sheep caused by the protozoan, *Theileria parva*.
- **Anaplasmosis** is a protozoan disease caused by *Anaplasma marginale*.
- **The Indian tick typhus** is caused by *Rickettsia conori* in cattle in UP and Assam.
- **Lyme disease** is also transmitted by ticks. **KFD (Kyasanur Forest Disease)** is a viral disease transmitted by ticks.

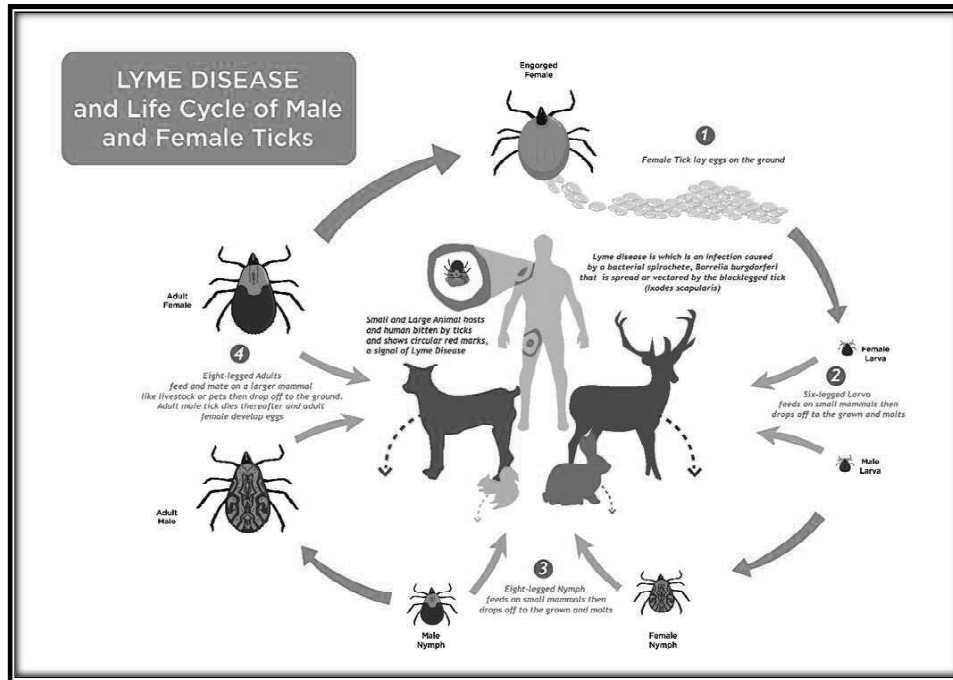


Fig. 4.19 Lyme Disease Caused by Ticks

Figure 4.19 is depicting the Lyme disease caused by ticks.

Control of ticks

Ticks can be controlled by hand-picking or by spraying lindane, carbaryl, malathion or arsenic trioxide.

(b) MITES as ectoparasite

A few ectoparasitic mites are as follows:

***Sarcoptes scabiei* – Mange mite**

Psoroptes equiovis*; *P. ovis*; *P. equi

- Mites can infect multiple hosts like man, horse, goat, sheep, cattle, dog etc.
- Mites are very tiny insect approximately 0.1 mm in size, oval in shape
- The colour of the mites varies from pale to greyish in colour.
- Adult possess 8 stubby legs
- Larvae are six legged.
- Eyes are absent.
- Mites possess piercing and sucking type of mouth parts.
- A few species of mite's burrow in skin and make galleries or wounds leading to intense itching.
- Later, fluid-filled pimples are formed which are covered with scabs.
- This fluid filled pimples contain abundant mites and are the chief sources of infection by contact.
- Host may lose hairs due to infection.

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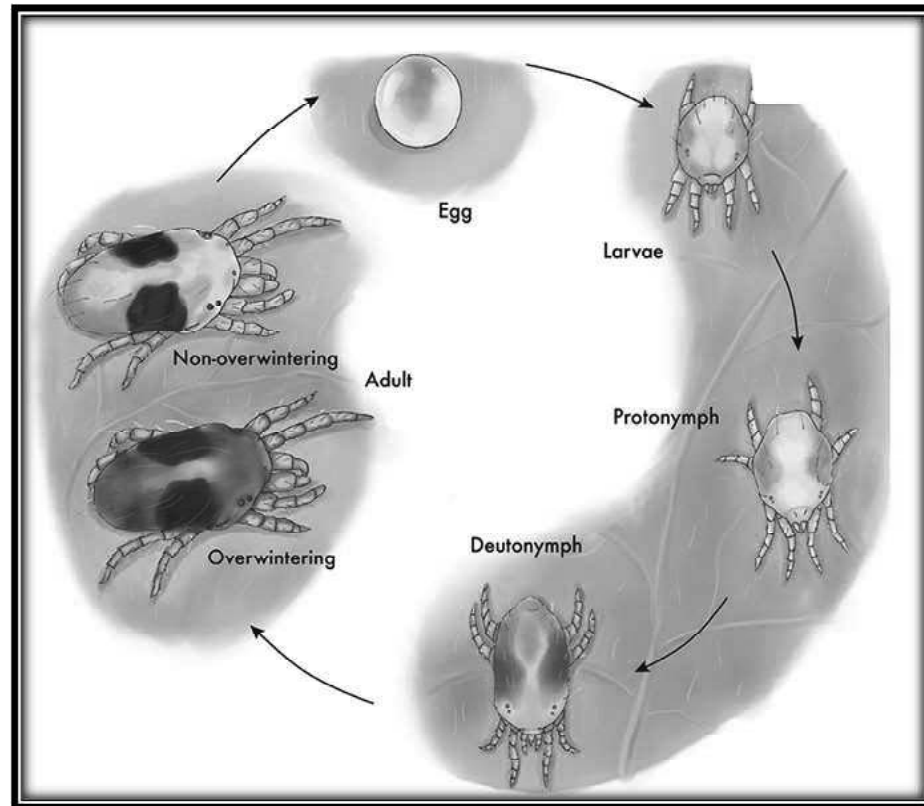


Fig.4.20 Life Cycle of Mites

Figure 4.20 is depicting the life cycle of mites

Control of Mites

Mites can be controlled by the application of Benzyl benzoate, Benzocaine-ethyl p-aminobenzoate, Srobiton mono-ocleate, Lindane and sulphur ointment or by giving bath with lime-sulphur, nicotine and lindane.

Check Your Progress

7. What do you mean by ectoparasites?
8. Trench fever is caused by which ectoparasite?
9. Which organism cause trench fever?
10. How does males and females Mosquitoes can be differentiated?
11. What are the four stages of life cycle if a mosquito?
12. How ticks can be controlled?

4.4 PARASITIC ADAPTATIONS IN PARASITES

Parasites abound with specialized characteristics associated with disease, and these are commonly interpreted as adaptations to the parasitic lifestyle. For example, a function we see as a 'derived' adaptation to parasitism may really have originated prior to parasitism; thus parasitism may either be irrelevant to the origin of that

function, or the origin of parasitism may have even been facilitated by that function.

The parasitic adaptation can be defined as the profound changes and modifications occurring in per-suit of successful living so that the parasite is fully adapted inside the body of the host.

For the most part, parasitic protozoans live in a fairly constant environment. Temperature fluctuates very little, or not at all, inside the host, desiccation is not a risk, and food is in constant supply. Free-living protists, on the other hand, face short- or long-term changes in temperature, aquatic acidity, food supply, moisture, and light. Many protozoans respond to adverse environmental conditions by encysting: they secrete a thick, tough wall around themselves and effectively enter a quiescent state comparable to hibernation. The ability to form a resistant cyst is widespread among diverse protistan groups and probably developed early in their evolutionary history. Resting cysts also are easily carried by the wind and form an important means of dispersal for species that live in the soil or are common in ephemeral ponds and pools. In climates with distinct cold seasons, the cyst may be an important phase in the annual life cycle.

The cyst wall is composed of a varying number of layers, the components of which are dependent on the species. During the encystment process, the protozoan cell undergoes a series of changes that considerably reduce the complexity of the organism. Flagellated organisms and ciliates lose their flagella and cilia, the contractile vacuole and food vacuoles disappear, and the distribution of organelles within the cell may be reorganized. In some species the cell volume reduces considerably. These changes are reversed during the process of excystment.

Certain marine planktonic tintinnids are programmed to break out of their cysts en masse at times of the year when the food supply is abundant. *Helicostomella subulata*, for example, excysts in June in temperate waters and becomes numerous from July through October. It encysts again in October, sinking to the sediments, where it remains until the following year. The cyst is a normal part of the annual life cycle, and even laboratory populations of this ciliate encyst at the same time as the natural population. This type of life strategy pattern has been demonstrated in several other ciliates and in some amoebae.

For soil-dwelling protozoans the cyst is an important refuge when soil moisture disappears or when soil water becomes frozen. In soils that are subject to freezing and periodic short-term thawing, the protozoans rapidly excyst, feed, and reproduce and then encyst again when soil water becomes temporarily unavailable to them.

The cyst plays an important role in the life cycles of several parasitic protozoans that have a free-living dispersal stage, such as *Entamoeba histolytica* and *Cryptosporidium*. The cysts are excreted in the host's feces and survive in water or soil. Humans are usually infected through drinking contaminated water or eating raw fruit and vegetables grown where human feces are used as fertilizer.

Some freshwater protozoans, especially the ciliates *Spirostomum*, *Loxodes*, and *Plagiopyla*, avoid unpleasant conditions, especially lack of oxygen, by abandoning their bottom-dwelling way of life and swimming upward to position themselves at a level where some oxygen is available but where they are not in

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direct competition with planktonic species. They remain there until oxygen again becomes available on the lake bottom, at which time they migrate downward.

The widespread occurrence of mixotrophy involving algal symbiosis and the retention and sequestration of the plastids of photosynthetic prey by planktonic protozoans is believed to be an adaptation to waters where food is limited. Ciliates that retain plastids appear to be far more common in waters where food is scarce than in productive waters. An inverse relationship exists between this form of mixotrophy and the productivity of the ecosystem.

The evolutionary success and ubiquitous distribution of parasitic nematodes is associated with elaborate physical and biochemical adaptations to parasitism: from a tough collagenous outer cuticle and sophisticated attachment and invasion apparatus such as hooks and suckers, to the expression of immunomodulatory molecules to impair host defense. Nematodes within the order Trichurata, including the genera *Trichinella*, *Capillaria*, and *Trichuris*, are a particularly successful group, parasitizing a wide range of hosts. For example, *Trichinella spiralis*, the causative agent of trichinosis, has the ability to infect all mammals and has a significant public health and economic impact. The Capillaridea contain a number of species that parasitize nearly all organs of all classes of vertebrates, some of which, e.g., *Capillaria philippinensis*, are often fatal if untreated.

4.5 EVOLUTION OF PARASITISM

Transitions from a free-living existence to a parasitic one probably outnumber any other type of major evolutionary shift in life history strategy. Adoption of a parasitic mode of life has occurred repeatedly and independently more than once in many groups. For example, extant species of parasitic nematodes originate from several distinct transitions to parasitism. The same is true for many other taxa, such as copepods or isopods. Among red algae alone, there have been over 100 separate switches to a parasitic existence. Thus, considering all eukaryotes, there have been several hundred independent transitions to parasitism over evolutionary time. Yet, despite their diverse origins and whether they have undergone extensive diversification or not, once they adopt parasitism as their mode of life, all these different lineages face the same set of selective pressures. They must all solve similar problems associated with host-to-host transmission, invasion of and survival within the host, and sustainable exploitation of host resources. There are only so many ways to achieve this successfully, and phylogenetically unrelated parasite lineages have therefore inevitably converged toward similar end-points in terms of their basic mode of. Only a limited set of trait combinations can allow the persistence of a parasite population in both the short and long terms, and thus natural selection has pushed unrelated lineages down shared evolutionary paths toward one of these combinations.

It is widely accepted that parasites undergo a rapid evolution, and they often have short generation times and large population sizes, parasites may evolve far more rapidly than their hosts. Attempts to understand parasite evolution, and the relevance of that evolution to disease, go back at least half a century to the first observations of drug resistance evolution in bacteria. Babayan et al. propose that

the life history of parasitic microfilarial worms shows evidence of adaptive plasticity. Specifically, they propose that worm development inside a mammalian host changes in response to the host's immunity, and that the parasite's response matches predictions from life history theory.

As a group, parasites are extraordinarily diverse. Even closely related parasites may behave very differently, infecting different host species, causing different pathologies, or infecting different tissues. For example, *Escherichia coli* bacteria, a typically harmless inhabitant of the human gut, can, in different forms, cause diarrhea, intestinal bleeding, urinary tract infections, kidney bleeding, meningitis, and other diseases. Underlying this diversity is evolution. Parasitism is inherent to life. Parasites are found in almost every organism of all existing species on earth. Since the beginning of life parasitism was adopted by protorganisms to multiply. Actually parasitism must have occurred at an early stage of evolution. All living organisms have a uniform biochemical composition that points to a common origin in a common ancestor that lived a billion years ago. Life on earth was only possible as a consequence of parasitism in what was still a molecular world. In fact, life appeared on earth around 4.4-3.8 billion years ago as a consequence of molecular parasitism and the present day life forms still display relics of these ancient associations in their genomes.

Later diversification of life forms and species radiation was also the consequence of these multiple associations. These ancient events represent the first steps towards a host-parasite way of life although still at the level of molecules. Associations recognized as a host-parasite systems existed since the very first gene associations, long before the appearance of the genetic code and gene translation Parasitism has been an important promoter of biodiversity, commencing at the molecular level, the subsequent advent of the cell, and then followed by the development of the intracellular environment as an ecological niche. Molecular parasitism is clearly exemplified by transposable elements of the genome. Indeed, the DNA sequences called transposable elements are actually recognized as molecular parasites Transposable elements occur in both prokaryotes and eukaryotes. Transposition is the insertion of an identical copy of the transposable element into a new genomic site of the host. These insertions can cause deletions, inversions, and chromosome fusions that result in considerable genome plasticity, thus contributing to biodiversity

The extra chromosomal transmissible genetic elements (viruses, plasmids and bacteriophages) are genome fragments that depend on the host cell to multiply and therefore they could not have preceded the cell itself. These elements could have been the origin of nuclear DNA as well as other cellular. The extra chromosomal genetic elements and the transposable elements represent relics of primitive molecular parasites. They played a very important role in the evolution of life forms since their inclusion in the hosts' genomes' cell promoted genetic diversity. Certainly many of the alterations they induced were deleterious, but a number of them resulted in advantages for their hosts. In viruses, genomic parasites can also be found and have been termed 'satellite' based on the satellite-like discovered by Kassanis (1962) in some cultures of tobacco necrosis virus. Some are relatively benign and seem to be well adapted to the host, but others can cause deleterious

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effects. Mayo et al. (1999) emphasized virus relationships. They stated that the evolutionary process that led to viruses is complex and that it is impossible to define when a molecule is a 'commensal', when it is a 'parasite' when it is a 'symbiont', and when it becomes a part of the genome of the virus.

Also, the prokaryotic cell exhibits clearly the parasitism that once was the causal effect of their origin, not only the nucleus but also the whole cell is a chymera, a polyheterogenic state derived from a long history of parasite associations. The association of microorganisms that resulted in mitochondria, chloroplasts and other organelles granted a significant increase in the complexity of the living organisms that resulted in the improvement of their capacity to occupy new ecological niches. Furthermore, the advent of the cell offered possible new niches for parasites. Species vary in the degree to which they are subject to parasitism. The perpetuation of life depends on the fine-tuning of the dialectic 'conservation versus change', where the parasite host ecological relationship plays an essential role.

Coexistence of terrestrial vertebrates and arthropods has been continuing over 200 million years; various forms of parasitism originated independently in various groups of arthropods during this period. The association of Acari and insects with nests and shelters of their hosts played the main role in the origin of parasitism in these major groups of arthropods. The primary step in the evolution of parasitism was the permanent habitation in nests and borrows of mammals and birds in Mesozoic era. The second step was a substitution of various forms of schizophagy by the regular feeding on products of vital activity and dead parts of host body. The next step was the feeding on various body parts of vertebrate hosts, namely skin, hair, feathers, external excreta, and drops of blood. The final step was the development of the ability to damage skin and suck out the blood of vertebrates. In some taxa of astigmatid mites the parasitism on birds originated from phoresy: hypopi (heteromorphous deutonymphs) obtained the ability to absorb the liquid nutrients from hair follicles and subcutaneous tissues through the cuticle. The development of haematophagous feeding on mammals in several families of Diptera was the second way of the origin of parasitism. Highly mobile dipterans with the piercing-sucking or licking mouthparts were able to change easily from the accidental puncturing of the host skin or licking of the blood, pus, and mucus to the obligatory haematophagy. The evolution of some arthropod taxa did not go beyond a primary domination of spatial relations, as in many astigmatid mites, or trophic relations in the form of micropredatory, as in the haematophagous Diptera.

Check Your Progress

13. How do protozoans respond to adverse environmental conditions?
14. List some physical and biochemical adaptations to parasitism shown by nematodes.
15. Why is parasitism considered as an important promoter of biodiversity?
16. What is transposition?

4.6 ANSWERS TO 'CHECK YOUR PROGRESS'

1. The majority of metazoan parasites of vertebrates are representatives of two phyla, which are as follows:
 - (i) Acoelomate platyhelminths
 - (ii) Pseudocoelomate nematoda
2. The filarial nematodes that can parasitize humans include the following:
 - *Wuchereria bancrofti*
 - *Brugia malayi*
 - *Brugia timori*
 - *Loa loa*
 - *Onchocerca volvulus*
 - *Mansonella perstans*
 - *Mansonella streptocerca*
 - *Dipetalonema streptocerca*
3. Cestodes are dorsoventral worms that are commonly known as tapeworms. They possess a segmented body that resembles a measuring tape and the term 'tapeworm' is thus used.
4. *Taenia saginata* is found in Africa, Eastern Europe, Southeast Asia and Latin America.
5. *Taenia saginata* has four cup-shaped suckers.
6. Usually, the CSF examination of *Taenia saginata* shows raised proteins, reduced sugar and pleocytosis.
7. Ectoparasites refers to the organisms that are found either on the skin or only in the superficial layers of the skin.
8. The ectoparasites that cause human disease fall into two main categories: insects (six-legged arthropods) and arachnids (eight-legged arthropods).
9. Trench fever is caused by *Rickettsia quintana*.
10. Males and females can be differentiated by the different structure of the antennae.
11. Mosquitoes are holometabolous insects showing all the four different stages, i.e., egg, larvae, pupa and adult.
12. Ticks can be controlled by hand-picking or by spraying lindane, carbaryl, malathion or arsenic trioxide.
13. Many protozoans respond to adverse environmental conditions by encysting: they secrete a thick, tough wall around themselves and effectively enter a quiescent state comparable to hibernation.
14. A tough collagenous outer cuticle and sophisticated attachment and invasion apparatus such as hooks and suckers and the expression of immunomodulatory molecules to impair host defense are some adaptations shown by nematodes.

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15. Parasitism has been an important promoter of biodiversity, commencing at the molecular level, the subsequent advent of the cell, and then followed by the development of the intracellular environment as an ecological niche.
16. Transposition is the insertion of an identical copy of the transposable element into a new genomic site of the host.

4.7 SUMMARY

- The word 'helminth' has been derived from the Greek word 'helmins' meaning worms. The helminthic parasites are multicellular and bilaterally symmetrical in appearance.
- Cestodes are tapeworms with segmented bodies that resemble a measuringtape.
- The length of a cestode ranges from a few millimetres to several metres.
- Intestinal helminths are a type of intestinal parasite that resides in the human gastrointestinal tract
- Intestinal helminths can be identified by fecal testing.
- An adult worm is cylindrical in shape and creamy white to pinkish in colour.
- Tapeworms are hermaphroditic, i.e., they have both testes and ovaries.
- Tapeworms lack a body cavity, mouth or digestive system and they absorb food through their body surface. They have a simple excretory system.
- The life span of an adult worm is about 365 days.
- A worm normally lives in the lumen of the small intestine and it feeds on the intestinal contents.
- Every female tapeworm can produce approximately 240,000 eggs per day, which are passed in the faeces.
- *Taenia saginata* is 4 to 6 meters long and 12 mm broad and it has a pear-shaped head.
- The diagnosis of *T. saginata* infection is made by the demonstration of proglottids or eggs in faeces.
- Intestinal helminths are a type of intestinal parasite that resides in the human gastrointestinal tract. They can be identified by fecal testing.
- The ectoparasites that cause human disease fall into two main categories: insects (six-legged arthropods) and arachnids (eight-legged arthropods).
- The body on insect is divided into three distinct regions: head, thorax and abdomen.
- Arachnids are a Class (Arachnida) of joint-legged Invertebrate Animals in the subphylum Chelicerata.
- Human lice are ubiquitous, i.e., widespread. The habitat of the human louse is solely on the human body or in the clothes.
- Relapsing fever is transmitted via spirochaet, *Borrelia recurrentis*.

- Fleas breed in dark and secluded places having high moisture contents like dusty floors, under carpets, rat burrows, granaries, chicken houses as well as godowns.
- Human flea is a cosmopolitan species, which is commonly found in hilly areas. Man is the preferred host, however, it can feed on other animals as well.
- Rat flea is a combless flea seen on rats and act as the vector of dreaded plague.
- Fleas majorly transmit plague that is caused by the safety pin bacillus called *Yersinia pestis* or *Pasturella pestis*.
- Mosquitoes are holometabolous insects showing all the four different stages i.e., egg, larvae, pupa and adult.
- Most of the common and important mosquitoes act as transmitters of pathogens for animal as well as man belong to four different genera namely: *Anopheles*, *Culex*, *Mansonia* and *Aedes*.
- There are more than 3000 species of mosquitoes in the world which are virtually distributed everywhere except in Iceland and poles.
- House fly is a larger insect, approximately 6-7 mm long, greyish in colour with blackish markings on the body.
- Pathogenic organism attaches to the lower side of oral disc, abdomen and hairs of tibia of housefly. These pathogenic organisms are carried to the food and water by housefly and finally enter into human system through contamination.
- Tsetse fly belongs to genus *Glossina* of family Glossinidae and is pronounced **tse-tse** or **teet-see** or **set-see**.
- Ticks are usually whitish to bluish-grey in colour having an oval body structure wrapped inside a tough and wrinkled skin.
- Ticks prefer to suck blood from soft parts of the host body, like ear pinna or base of the tail.
- Ticks can be controlled by hand-picking or by spraying lindane, carbaryl, malathion or arsenic trioxide.
- Parasites abound with specialized characteristics associated with disease, and these are commonly interpreted as adaptations to the parasitic lifestyle.
- Parasitic adaptation can be defined as the profound changes and modifications occurring in per-suit of successful living.
- Parasites played a very important role in the evolution of life forms since their inclusion in the hosts' genomes' cell promoted genetic diversity.

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4.8 KEY TERMS

- **Cestode:** It is a dorsoventral tapeworm with a segmented body.
- **Lumen:** It is the cavity through which digested food passes and from where nutrients are absorbed.

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- **Oncosphere:** It is the larvae of the tapeworm contained within the external embryonic envelope and armed with six hooks.
- **CSF:** Short for cerebro spinal fluid, it is a clear fluid that occupies the sub-arachnoid space and the ventricular system around and inside the brain and spinal cord.
- **Ectoparasites:** Ectoparasites refers to the organisms that are found either on the skin or only in the superficial layers of the skin.
- **Biological transmission:** The disease-causing pathogens are carried inside the system of the insect. The infectious agent undergoes development or multiplication inside the vector. This is referred to as biological transmission of the infectious agent by the vector.
- **Trench fever:** Trench fever is caused by *Rickettsia quintana*. This disease was prominent among soldiers during the World War II.
- **Parasitic Adaptation:** The parasitic adaptation can be defined as the profound changes and modifications occurring in per-suit of successful living so that the parasite is fully adapted inside the body of the host.

4.9 SELF-ASSESSMENT QUESTIONS AND EXERCISES

Short-Answer Questions

1. What are the types of nematods?
2. What is the specialty of a cestode?
3. How do you distinguish between the different species of *T. saginata*?
4. When does *T. saginata* occur?
5. What are the guidelines for identification of intestinal helminthes?
6. How does insect acts as vector of disease?
7. How could we control of bedbugs?
8. What is the treatment given to the patient of malaria?
9. Name some disease transmitted by tick infection.
10. What are the three stages of fever in malaria?
11. How cyst plays an important role in the life cycles of several parasitic protozoans
12. How parasites did played a very important role in the evolution of life forms?

Long-Answer Questions

1. Explain the morphology, life cycle and lab diagnosis of *A. Lumbricoides*.
2. Describe with a diagram the morphology and life cycle of *T. saginata*.
3. Explain the lab diagnosis of *T. saginata* infection in humans.

4. Describe the life cycle of *T. solium*.
5. Analyze the generalized life cycle of *Pediculus humanus*.
6. Explain the symptoms and pathogenesis of malaria.
7. Elaborate on the parasitic adaptations in parasites.
8. Write a brief note on the evolution of parasitism.

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4.10 FURTHER READING

- Pawar, Kishore R, Ashok E Desai. 2016. *Parasitology*. New Delhi: Nirali Prakashan.
- Heelan, Judith Stephenson, Frances W. Ingersoll. 2002. *Essentials of Human Parasitology*. US: Delmar publishers
- Sood, Ramnik. 2019. *Textbook of Human Parasitology Protozoology and Helminthology*. New Delhi: CBS PUB & DIST PVT Limited INDIA.
- Myerowitz, Richard L. 1983. *Zygomycosis (mucormycosis). The Pathology of Opportunistic Infections with Pathogenetic, Diagnostic, and Clinical Correlations*. New York: Raven Press.
- John, David T. 2006. , et al. *Markell and Voge's Medical Parasitology*, 9th edition. St. Louis, MO: Saunders Elsevier.

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