NATIONAL OPEN UNIVERSITY OF NIGERIA

BIO 217

General Microbiology

Module 5

BIO 217 (General Microbiology)
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Module 5

Unit 1 Mechanisms of Genetic Variation and Hereditary

1.0 Introduction

Each living organism resembles its ancestors in most of its characters. The maintenance of specific properties, that is, the constancy of characters over generations is called heredity.

Genetics is the study of inheritance (heredity) and the variability of the characteristics of an organism. Inheritance exacts transmission of genetic information from parents to their progeny (offspring). Deoxyribonucleic acid (DNA) is the chemical substance responsible for hereditary in all cells because it beams the genetic information. Each genetic character can be assigned to a gene which carries the information. Microorganisms are capable of transmitting genetic information from generation to generation with great accuracy. (This unit examines the causes of variation in microorganisms). Variability or variation of the inherited characteristics occurs or alters as a result of change in the genetic makeup of a cell or in environmental conditions. Variation in microorganisms takes place by mutation. Recombination and gene transfer.

2.0 Objectives

At the end of this unit, you should be able to:

- define the term genetic variation
- Define mutation.

3.0 Main Content

Genetic Variation
This is changes in or of a gene which leads to a loss of the enzymes or to the production of an altered enzymes, hence, to recognizable changes in the hereditary character.

Genetic variation in bacteria can take place by:

- mutation
- gene transfer or recombination.

3.1 Mutation

Mutation can be defined as a change in the nucleotide sequence of DNA. Mutation can involve the addition, deletion or substitution of nucleotides. These changes in the nucleotides are stable and heritable and are passed down from one generation to the next. Mutation introduces genetic variation among organisms.

A mutant is a strain of any cell or virus carrying a change in the nucleotide sequence.

A mutant is different from its parent strain in:

Genotype: The nucleotide sequence of the genome.

The Phenotype: The observable property of the mutant in the altered phenotype is called a mutant phenotype.

The strain isolated originally from nature is called the wild type strain.

Mutation can occur spontaneously or induced under the influence of external agents (mutagens).

3.1.1 Spontaneous Mutation

This is mutation that occurs without exposure to external agents or any known mutagenic treatment. It occurs at a fairly constant frequency in a particular organism, one per 10^6 to 10^{10} in a population derived from a single bacterium. It may result from errors in DNA replication or from the action of mobile genetic molecules called transposons.

Mutation arising from error in DNA replication can be:

- Transition Mutation: This is the substitution of a purine for another purine (A or G) or one pyrimidine for another pyrimidine (C or T) that lead to a stable alteration of the nucleotide sequence. This type of mutation is relatively common.
- Transversion Mutation: This is a mutation in which a purine is substited for a pyrimidine or pyrimidine for a purine. This is rarer due to the steric problems of pairing purines with purines and pyrimidine with pyrimidine. Both Transition and Transversion mutations are types of base substitution in point mutation.
- Mutations as result of lesions on DNA which result in apurine sites, apyrimidinic sites, oxidation of DNA.
- Mutation as a result of the insertion of DNA segments into genes. Insertion usually inactivates genes. They are caused by movement of insertion sequences and transpons.

3.1.2 Induced Mutation

This is mutation caused by external agent (mutagens), which may be physical or chemical agents.

- **Physical Agents**: Physical agents include ultraviolet (UV) light, raising radiation, visible light and heat. Ionizing radiation (e.g. X-rays) can break both single and double strand DNA. The frequency of mutation is greatly increased at a temperature of 37°C and above.
- **Chemical Agent**: There are three main types of mutagenic chemicals.
- **Base analogs**: These are chemicals structurally similar to normal DNA bases and can be substituted for them during DNA replication. These compounds exhibit based pairing properties different from the bases they replace and eventually cause a stable mutation. An example: 5-bromouracil (5-BU) an analog of thymine.

- **DNA Modifying Agents**: These chemicals react chemically with DNA. They change a base structure and alter its base pairing characteristics. Examples are Methyl-nitrosoguanidine and Hydroxylamine.
- **Intercalating Agents**: These are chemicals with flat molecules that can intercalate (slip in) between bases pairs in the central stack of the DNA helix. They include single nucleotide pair insertion and deletions. Examples are acridine such as proflavin and acridine orange.

3.2 Types of Mutation

Two common types of mutation are: point mutation and frame shift mutation.

Point Mutations
Point mutations occur as a result of the substitution of one nucleotide for another in the specific nucleotide sequence of a gene or defined as change in only one base pair.

Point mutations in protein – coding genes can affect protein structure and are named according to if and how they change the encoded protein.

The most common type of point mutations are:

Silent mutation: Change the nucleotide sequence of a codon but do not change the amino acid encoded by that codon.

Missence Mutations: This involves a single base substitution that changes a codon for one amino acid into a codon for another.

Nonsense Mutation: This causes the early termination of translation and therefore results in a shortened polypeptide. They are called nonsense mutation because they convert a sense codon to a non-sense or stop code.

Frame Shift Mutations: These mutations result form an addition or loss of one or more nucleotides in a gene and are termed insertion or deletion mutations respectively.

This results in a shift or the reading frame.
Frame shift mutations usually are very harmful and yield mutant phenotypes resulting from the synthesis of non-functional protein.

The effect of mutation can be described at the protein level and in terms of traits or other easily observed phenotypes.

A mutation from wild type to a mutant form is called a forward mutation; however, a reversion mutation occurs when second mutual at the same site as the original mutation which restore the wild type phenotype. If the second mutation is at a different site than the original mutation is called suppressor mutation.

Self-Assessment Exercise

i. What are point mutations?

ii. What are frame shift mutations?

3.3 Genetic Recombination

This is the formation of a new genotype by reassortment of genes following an exchange of genetic material between two different chromosomes which have similar gene at corresponding sites and are from different individuals. Progeny or offspring from recombination have combination of genes different from those that are present in the parents. In bacteria, genetic recombination's result from three types of gene transfer, they are:

Conjugation: This is the transfer of genes between cells that are in physical contact with one another.

Transduction: This is the transfer of genes from one cell to another by a bacteriophage.

Transformation: This is the transfer of cell free or naked DNA from one cell to another.

3.4 Mechanism of Recombination

Inside the recipient cell, the donor DNA fragment is positioned alongside the recipient DNA in such a way that homologous genes are adjacent. Enzymes act on the recipient DNA, causing nic and excision of a fragment. The donor is then integrated into the recipient chromosome in place of the excised DNA. The recipient becomes the combination cell because its chromosomes contain DNA of both the donor and the recipient cell.

3.4.1 Conjugation

This is a mechanism of genetic transfer that involves cell to cell contact. It is plasmid encoded mechanism i.e. it is controlled by gene carried by certain plasmid (such as F plasmid). The process of conjugation involves a donor cell which contains conjugative plasmid and a recipient cell which does not.

F Plasmid
The F plasmid (F stands for fertility) is a circular DNA molecule of 99159 bp. It is an extra chromosome DNA that encodes the necessary information necessary for conjugation. The F plasmid has a large region of DNA, the extra region containing genes that encode transfer functions. Many genes in the extra region are involved in meeting pair formation and most of these have to do with the synthesis of a surface structure the sex pilus (plural, pili).

Only donor cells produce these pili. Pili allow specific pairing to take place between the donor and recipient cells. The pilus makes specific contact with a receptor on the recipient cell and is retracted by disassembling its subunit. This pulls the two cells together, making the donor and recipient cells remain in contact by binding proteins located in the outer membrane of each cell DNA is the transfer from donor to recipient cells through this conjugation junction.

Mechanisms of DNA Transfer
During conjugation DNA transfer is triggered by cell to cell contact by which one strand of the circular plasmid is mixed and is transferred to the recipient. The nicking enzyme required to initiate the process that is encoded by the of the F plasmid DNA synthesis by the rolling circle mechanisms replaces the transferred strand in the donor while a complementary DNA strand is being made in the recipient. At the end of the process, both donor and recipient cells possess complete plasmid. For transfer of the F plasmid, if an F-

containing donor cell (designated as F^+) mates with a recipient cell lacking the plasmid (designated as F^-) the result is two F^+ cells.

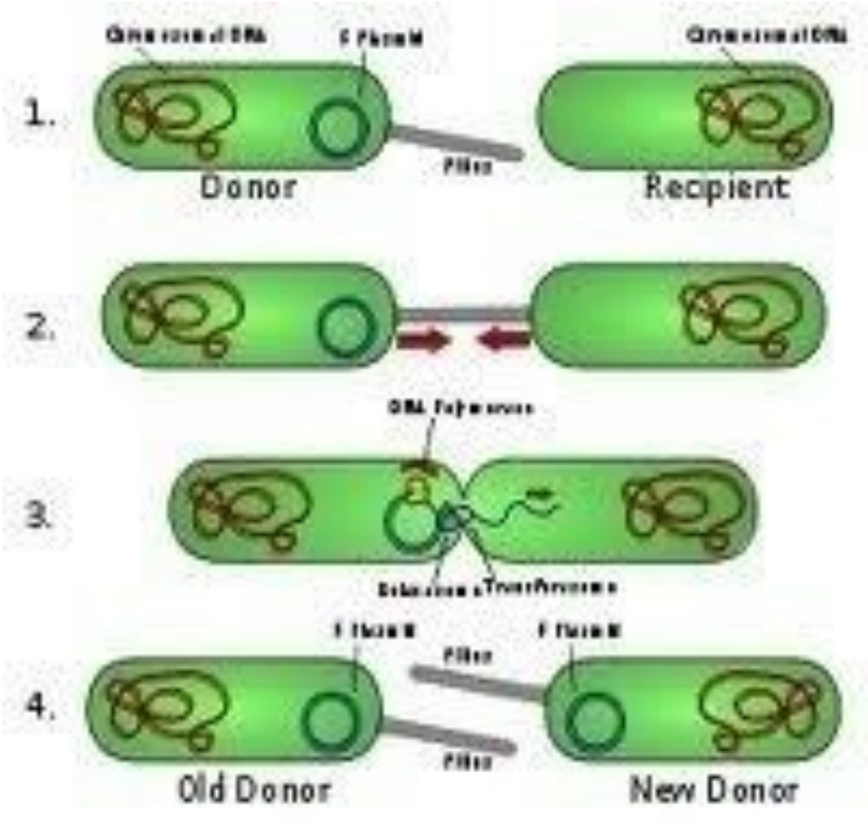
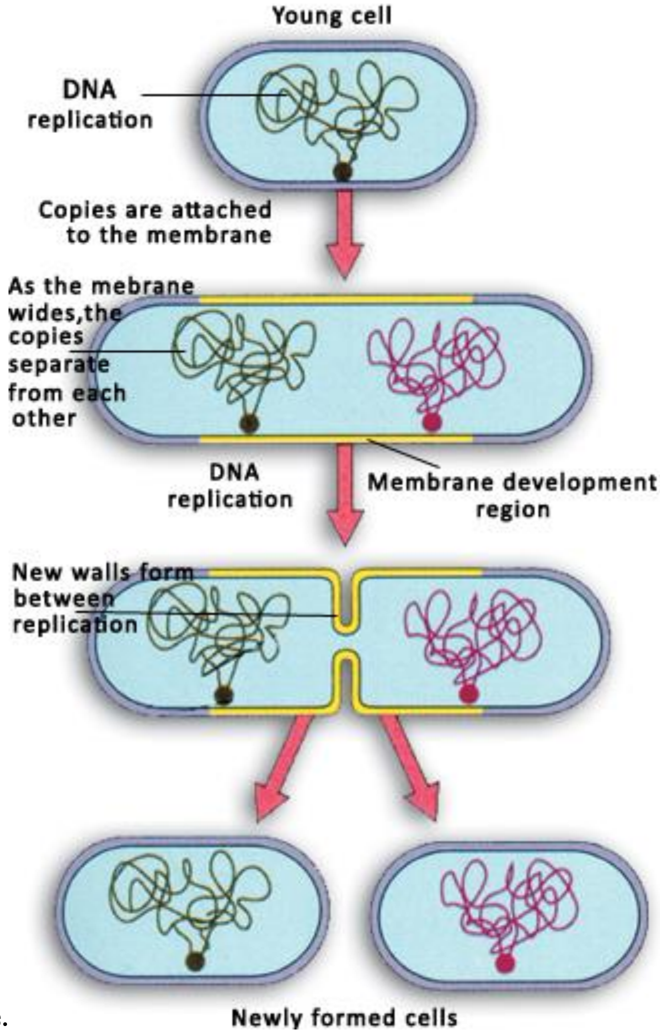


Fig. 1: The Diagrammatic Illustration of Conjugation Source

Source: Answers.com

3.4.2 Transformation

This is a genetic transfer process by which free DNA is incorporated into a recipient cell and brings about genetic change. Several prokaryotes are naturally transformed including certain species of Gram negative and Gram positive bacteria and some species of Archaea because the DNA of prokaryotes is present in the cell as a large single molecule, when the cell is gently lysed, the DNA pours out and breaks easily into fragments containing genes which are released into the surrounding environment. If a fragment contacts a competent cell, a cell that is able to take up DNA and be transformed, the DNA can be bound to the



cell and taken inside.

Fig. 2: The Diagrammatic Illustration of Transformation in Cells

Source: harunyahya.com

Competency for transformation is a complex phenomenon and is dependent on several conditions such as stage of growth. Natural transformation has been discovered so far in certain genera including *Streptococcus, Bacillus, Acinectobacter, Azobacter, Helicobacter* and *Pseudomonas*. Gene transfer by this in process occurs in soil and aquatic environment.

3.4.3 Transduction

This is a mechanism of genetic transfer in which a bacterial virus (bacteriophage) transfers DNA from one cell to another. Virus can transfer the host genes in two ways:

- generalised transduction and
- specialised transduction

Generalised Transduction

DNA derived from virtually any portion of the host genome is packaged inside the mature union in place of the virus genome. Any gene on the donor chromosome can be transferred to the recipient since they carry any of the host chromosome, they are called generalised transduction. When a bacteria cell is infected with a phage, the lytic cycle is initiated. During the lytic infection, the enzymes responsible for packaging viral DNA into the bacteriophage sometimes package host DNA accidentally. The resulting union is called a transducing particle. On lysis of the host cells, the transducing particles are released along with normal union (that is those containing the virus genome), hence the lysate is used to infect a population of recipient cells.

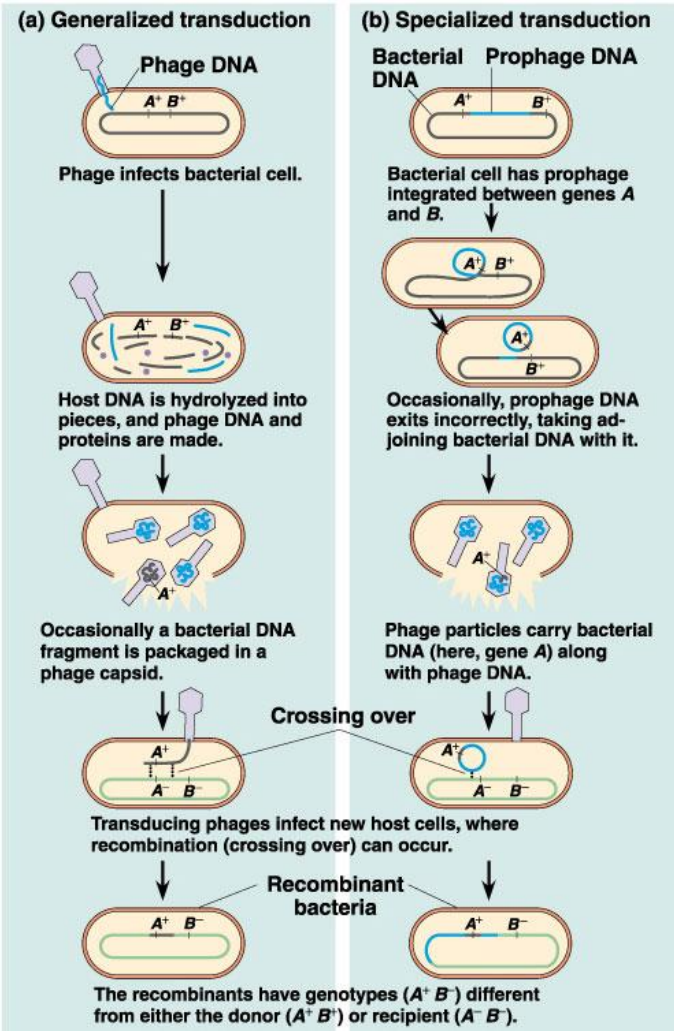


Fig. 3: The Diagrammatic Illustration of Generalised Transduction

Source: bio.miami.edu

Most of the cells become infected with normal virus. However, a small portion of the population receives transducing particles that inject the DNA they packaged from the previous host bacterium. These transducing particles undergo genetic recombination with the DNA of the new host.

Generalised transduction allows the transfer of any gene from one bacterium to another at a low frequency.

Specialised Transduction: In specialised transduction, DNA from a specific region of the host chromosome is integrated directly into the virus genome usually replacing some of the virus genes. This occurs only in a certain temperate viruses. Specialised transduction allows extremely efficient transfer but is selective and transfers only a small region of the bacteria chromosome. In the first case of specialised transduction to be discovered, gelatose genes were translated by phage lambda of *E. Coli*.

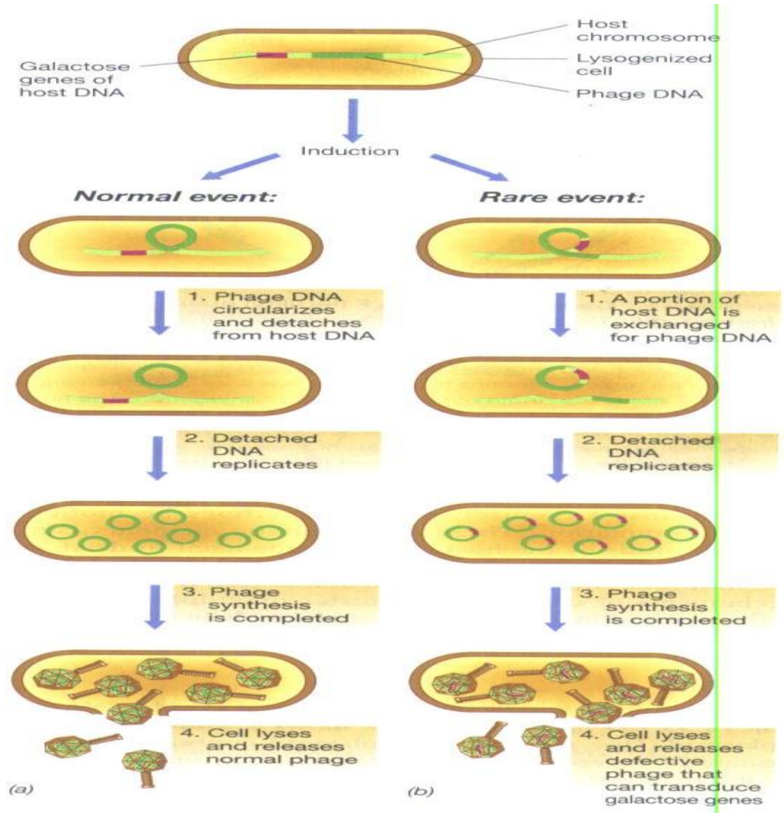


Fig. 4: The Diagrammatic Illustration of Specialised Transduction

Source: Brock's *Biology of Microorganisms* by Medigan *et al*., (2009)

When lambda lysogenizes a host cell, phage genomes are integrated into the host DNA at a specific site. The region in which lambda integrates in the *E. Coli* chromosome is next to the cluster of the gene that encode the enzyme for galactose utilisation. After insertion, viral DNA replication is under control of bacterial host chromosome. Upon induction, the viral DNA separates from the host DNA by a process that is reverse of integration.

The phenotype of microorganisms can be affected in various ways:

i. Morphological Mutations: Changes in the microorganism colonial or cellular morphology of the microorganisms.

ii. Lethal Mutation: Resulting in the death of the microorganism.

iii. Biochemical Mutation: Causing a change in the biochemistry of the cell.

3.4.4 Mutation Detection involves

- Visual Observation
- Replace Plating Technique.

4.0 Conclusion

Microbial variation can arise as a result of mutations which can occur spontaneous or induced by physical and chemical changes. Mutation leads to changes in protein structure and function which in turn alter the phenotype of an organism. This results in the organism (mutant) being different from the one found originally in nature (the wild type).

5.0 Summary

- Mutation is a stable heritable change in the nucleotide sequence of a DNA molecule.
- Mutation can be spontaneous or induced.
- Spontaneous mutation can arise from replication errors (transition, transversion, addition and deletion of nucleotide) from DNA lesions and from insertion of DNA segments and transpons.
- Induced mutation can be from physical and chemical agents called mutagens.
- Physical agents, of mutation include ultraviolet rays, ionizing rays, radiation, light, heat, etc.
- Chemical agents of mutation are base analogs, intercalating agents and DNA modifying agents.
- Main types of mutation are point mutation and frame shift mutation.
- Mutations are recognised when they cause a change from the normal wild type phenotype. A mutant phenotype can be restored to wild type by either reversion or suppressor mutations.
- Some major types of mutations categorised based on the effect on phenotypes are morphological, lethal and biochemical.
- Replace plating can be used for detection and isolating mutants.
- Amen test is used to screen for mutagens and potential carcinogens.

6.0 Self-Assessment Exercise

1. What is mutation?
2. List four ways in which spontaneous mutation might arise.
3. List three methods of recombination in a prokaryotic cell.
4. Explain the mechanism of conjugation in bacteria.

7.0 References/Further Reading

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Unit 2 Biogeochemical Cycling Of Elements

1.0 Introduction

Life on earth would not be possible without microbes. Soil microorganisms serve as biogeochemical agents for the conversion of complex organic compounds into simple inorganic compounds or into their constituents' elements. The overall process is called Mineralisation. Biogeochemical cycling refers to the biological and chemical processes that elements such as carbon, nitrogen, sulfur, iron and magnesium undergo during microbial metabolism. This conversion of complex organic compounds into inorganic compounds or elements provides for the continuity of elements (or their compounds) as nutrients for plants and animals including man. This unit examines the biogeochemical cycles of elements such as carbon, sulfur, nitrogen and phosphorous.

2.0 Objectives

At the end of this unit, you should be able to:

- define biogeochemical cycling
- state the features of biogeochemical cycles
- describe the carbon cycle
- describe the sulphur cycle
- Describe the phosphorus cycle.

3.0 Main Content

Biogeochemical Cycling
Biogeochemical cycling is the movement of materials via biochemical reactions through biospheres. The biosphere is that portion of the earth and its atmosphere in which living organisms occur. The activities of microorganisms within the biosphere have a direct impact on the quality of human life. Microorganisms are especially important in recycling materials.

The metabolism carried out by microorganisms often transfers materials from one place to another. Changes in the chemical forms of various elements can lead to the physical movement of materials. Sometimes causing transfer between the atmosphere (air) hydrosphere (water) and lithosphere (land).

Biogeochemical cycling also refers to the biological and chemical processes that elements such as carbon, nitrogen, sulfur, iron and magnesium undergo during microbial metabolism.

It can also be defined as cyclical path that elements take as they flow through living (biotic) and non-living (abiotic) components of the ecosystem. These cycles are important because a fixed and limited amount of the elements that make up living cells exists on earth and in the atmosphere. Thus in order for an ecosystem to maintain and sustain its characteristics and life forms elements must continuously be recycled. For example, if the organic carbon that animals use as an energy source and exhale as carbon dioxide (CO_2) were not eventually converted back to an organic form we would run out of organic carbon to build cells.

Elements involved in the biogeochemical cycles are used for three general purposes.

Biomass Production: In biomass production, the element transferred (e.g. N, C, etc) is incorporated into cell materials for example, all organisms require nitrogen to produce amino acids, hence plants and many prokaryotes assimilates nitrogen by incorporating ammonia (NH_3) to synthesise the amino acid glutamate. Animals cannot incorporate ammonia instead require amino acids in their diets. Some prokaryotes can reduce atmosphere nitrogen to form ammonia, which can then be incorporated into cellular material.

Energy Source: A reduced form of the element is used to generate energy in form of ATP. For example reduced carbon compounds such as lipids and amino acids are used as energy source by heterotrophs. Chemolitrophs can use reduced inorganic molecules such as hydrogen sulfide (H_2S) ammonia (NH_3) and Hydrogen gas.

Terminal Electron Acceptor i.e. Carbohydrate Oxidised to CO_2: Electrons from the energy source are transferred to an oxidised form of the element during respiration, this reduces the terminal electron acceptor in aerobic conditions, O_2 is used as a terminal electron acceptor. In anaerobic conditions, some prokaryotes can use nitrate, sulphate (SO_4), and carbon dioxide (CO_2) as terminal electron acceptors.

Global climate change, temperature precipitation and stability of ecosystem are all dependent on biogeochemical cycles.

3.1 Peculiar Features of Biogeochemical Cycles

- Elements required are in five forms and mostly from non-living reservoir in the atmosphere. They can also be gotten from sedimentary rock or water.
- The elements go in cycle and are always free in inorganic state in abiotic environment and when needed in biotic environment, they are turned to organic state. They can be combined with other elements.
- The recycling of these elements maintains a necessary balance of nutrient and they are maintained throughout.
- The cycles (biogeochemical) are complex and they involve the activity of producers, consumers and decomposers.
- All organisms participate directly in recycling by removing, adding or altering nutrients. Microorganisms are noted for metabolic conversion especially in changing some elements from one nutritional form to another.
- The total turnover rate of element is most rapid in atmosphere and lowest in sedimentary rocks.

Self-Assessment Exercise

- What is biogeochemical cycle of elements?
- State three features of biogeochemical cycles.

3.2 Carbon Cycle

Carbon is a very important element as it makes up organic matter which is a part of all life. Carbon follows a certain route on earth called the CARBON CYCLE.

The carbon cycle primarily involves the transfer of carbon dioxide and organic carbon between the atmosphere where carbon occurs principally as inorganic CO_2 and the hydrosphere and lithosphere which contain varying concentrations of organic and inorganic compounds.

The carbon cycle begins with carbon fixation, which is the conversion of CO_2 to organic matter. Plants are thought of as the principal CO_2 fixing organisms but at least half of the carbon on earth is fixed by microbes; particularly marine photosynthetic prokaryotes and protists.

Cyanobacteria such as *Prochlorococcus* and *Synechococcus* are involved in carbon fixation using energy from sunlight. Chemolithoautotrophic microorganisms such as *Thiobacillus* and *Beggiatoa* also fix CO_2 into organic matter while metabolising compounds such as H_2S for energy.

Once carbon is fixed into organic compounds, the next stage in the cycle involves its transfer from population to population within the biological community, supporting the growth of a variety of heterotrophic organisms, i.e. heterotrophs such as animals and protozoa that eat autotrophs and may in turn be eaten by other animals. Hence, they acquire organic carbon to build biomass and to oxidize to gain energy. Decomposers use the remains of primary producers and consumers for the same purposes.

The respiratory and fermentative metabolism of heterotrophic organisms returns inorganic carbon dioxide to the atmosphere completing the carbon cycle. When plants and animals die, these organic compounds are decomposed by bacteria and fungi and during decomposition the organic compounds are oxidised and CO_2 is returned to the cycle.

Carbon is stored in rocks such as limestone ($CaCO_3$) and is dissolved as carbonate ions (CO_3) in oceans. Vast deposits of fossil organic matter exist in the form of fossil fuel such as coal and petroleum. Burning these fuel releases CO_2 resulting in an increased amount of CO_2 in the atmosphere.

Alternatively, inorganic CO_2 and organic carbon can be reduced anaerobically to methane ((CH_4). Methane is produced by Archaea in anoxic habitats. Archaea such as *Methanobrevibacter* in the gut of termites also contribute to methane production.

The carbon cycle has come under intense scrutiny in the last decade. This is because CO_2 levels in the atmosphere have risen from their preindustrial concentration of about 280μmol per mol to 376μmol per mol in 2003. This represents an increase of about one-third and CO_2 levels continue to rise. Like CO_2, methane is also a greenhouse gas and its atmospheric concentration is likewise increasing about 1% per year, from 0.7 to 1.7ppm (volume) since the early 1700s. These changes are clearly the results of the combustion of fossil fuels and altered land use.

The term greenhouse gas describes the ability of these gases to trap heat within earth's atmosphere, leading to a documented increase in the planet's mean temperature. Indeed, over the past 100 years, earth's average temperature has increased by 0.6% and continues to rise at a rapid rate, i.e. global warming of the earth.

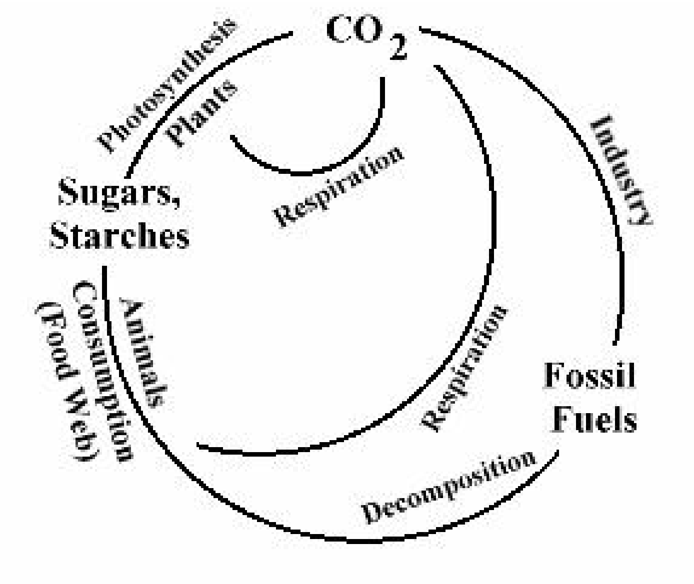

Fig. 1: Carbon Cycle

3.3 Nitrogen Cycle

Nitrogen is an essential component of DNA, RNA and proteins, which are the building blocks of life, hence all organisms require nitrogen to live and grow.

Nitrogen, the most abundant substance in the atmosphere or air (almost 80%) is not directly useable by most organisms. This is because the strong triple bond between the nitrogen atoms in the molecule makes it relatively inert. Only a few bacteria are able to utilise nitrogen directly. For plants and animals to be able to use N_2, N_2 gas must first be converted to more chemically available forms such as ammonium (NH_4^+), Nitrate (NO_3^{-1}), (NO_2^{-1}) and organic nitrogen containing compounds such as amino acids and proteins. Microorganisms also are able to utilise these other forms of nitrogen mentioned above.

The conversion of nitrogen compounds primarily by microorganisms changes the oxidation states of nitrogenous compounds and establishes a nitrogen cycle.

Three processes carried out by microorganisms are critical in the nitrogen cycle. They are:

1. nitrogen fixation

2. nitrification and
3. denitrification.

3.3.1 Nitrogen Fixation

This is strictly a bacterial process in which molecular nitrogen is converted to ammonium ion and it is the only naturally occurring process that makes nitrogen available to living organisms. It is carried out by a few bacteria that have a nitrogenase enzymes system.

This process brings nitrogen from the atmosphere to the hydrosphere and lithosphere. Because plants depend on the availability of nitrogen for growth, microbial metabolism of nitrogen containing compounds has a dramatic impact on agricultural productivity. Plants and animals rely entirely on the fixed forms of nitrogen (such as ammonium and nitrate ions) provided by bacterial nitrogen fixation. Nitrogen fixation involves two types of bacteria or microorganisms).

Free Living Nitrogen Fixing Bacteria: These bacteria are found in high numbers or concentration in the rhizosphere (the region where the soil and roots make contacts). In aquatic environment, blue green alga is an important free-living nitrogen fixer. Trichodesmium fix nitrogen aerobically while free-living anaerobes such as members of the genus *Clostridium* fix nitrogen anaerobically.

Symbiotic Nitrogen Fixing Bacteria: Members of the genera *Rhizobium*, *Bradyrhizobium* and others are important nitrogen fixing bacteria. They play an important role in plant growth for crop production. These bacteria live in association with leguminous plants. The bacteria invade the root cells to form nodules, receives carbohydrate and a favourable environment from their host plants in exchange for the nitrogen they fix. However, other bacterial symbiotics fix nitrogen, for example the *Actinomycete Frankia* fixes nitrogen while colonizing many type of woody shrub. The heterocystous cyanobacterium *Anabaenea* fixes nitrogen when in association with the water fern *Azolla*.

The product of nitrogen fixation is ammonia (NH_3). It is immediately incorporated into amino N-atoms and eventually as amino acids. The nitrogenase enzymes are very sensitive to oxygen and must be protected from oxidizing effects.

The nitrogen cycle continues with the degradation of these molecules into ammonium (NH_4^+) within mixed assemblages of microbes *Rhizobium* and *Bradyrhizobium* species generally exhibits rates of nitrogen fixation that are two or three times higher than these accompanied by free nitrogen fixing bacteria.

3.3.2 Nitrification

This is a process carried out by chemolithotrophic bacteria which convert ammonium ions to nitrate (NO_3^-) ions. It is a two step process whereby ammonium ion is first oxidised to nitrite (NO_2^-) which is then oxidised to nitrate.

First step $\longrightarrow$ NH_4^+
NO_2^-

Ammonium ion
Nitrite ion

Second step $\longrightarrow$ NO_2^-
NO_3^-

Nitrite ion
Nitrate ion

Bacteria of the genera *Nitrosomonas* and *Nitrosococcus* play important role in the first step. *Nitrobacter* and related *Chemolithotrophic* bacteria carry out the second step. In addition *Nitrosomonas eutropha* has been found to oxidise ammonium ion anaerobically to nitrite oxide (N.O) using nitrogen dioxide (NO_2) as an acceptor in a denitrification related reaction.

Genera of Nitrification Bacteria

Genus	Converts	Habitat
Nitrosomonas	Ammonia to nitrite	Soil, freshwater marine
Nitrosospira	Ammonia to nitrite	Soils
Nitrosococcus	Ammonia to nitrite	Soils, freshwater marine
Nitrosotobus	Ammonia to nitrite	Soils
Nitrobacter	Nitrite to nitrate	Soils, freshwater marine
Nitrospira	Nitrite to nitrate	Marine
Nitrococcus	Nitrite to nitrate	Marine

The production of nitrate is important because it can be reduced and incorporated into organic nitrogen. This process is known as assimilatory nitrate reduction (i.e. the use of nitrate as a source of organic nitrogen is an example of assimilatory reduction) because assimilatory reduction of nitrate sometimes accumulates a transient intermediate. Alternatively, some microorganisms use nitrate as a terminal electron acceptor during anaerobic respiration this is a form of dissimilatory reduction.

3.3.3 Denitrification

This is a process in which nitrate is removed from the ecosystem and returned to the atmosphere as dinitrogen gas (N_2) through a series of reactions. Denitrification leads to the return or loss of nitrogen to the atmosphere as nitrogen gas. Through this process, oxidised forms of nitrogen such as nitrate and nitrite are converted to dinitrogen (N_2) and to a lesser extent, nitrous oxide gas.

Denitrification is an anaerobic process carried out by denitrifying bacteria which convert nitrate to dinitrogen in the following sequence:

NO_3^- $\longrightarrow$ $\longrightarrow$ $\longrightarrow$ $\longrightarrow$ NO_2^-
NO N_2O N_2

Nitrate Nitrite
Nitric Nitrous
Nitrogen

Ion ion
oxide oxide gas

Nitric oxide and nitrous oxide are both environmentally important gases. N_2O is an important Greenhouse gas contributing to global climate change while nitric oxide contributes to smog. Once converted to dinitrogen nitrogen is unlikely to be reconverted to a biologically available form because it is a gas and is rapidly lost to the atmosphere.

Denitrification is the only nitrogen transformation that removes nitrogen from ecosystem (essentially irreversibly). Denitrification is a form of dissimilatory reduction. Finally, nitrate can be transformed to ammonia in dissimilatory reduction by a variety of bacteria, including *Geobacter, Metallireducens, Desulfovibro spp* and *Clostridium spp*. Other sp capable of transforming NO_3 to N_2 are *Achomobacter, Agrobacterium, Alkaligenes, Bacillus, Chromobacterium, Flavobacterium, Hypnomicrobium, Pseudomonas Thiobacillus* and *Vibrio*. From agricultural standpoint, it is an undesirable process because it leads to loss of nitrogen from the soil, hence a decline in nutrients for plant growth.

3.3.4 Ammonification

Ammonification is the decomposition process that converts organic nitrogen into ammonia (NH_3). A wide variety of organisms, including aerobic and anaerobic bacteria as well as fungi can degrade protein, this they do through the action of extracellular proteolytic enzymes that break down protein into short peptides or amino acids. After transport of the breakdown products into the cell, releasing ammonium, the decomposer will assimilate much of this compound to create biomass.

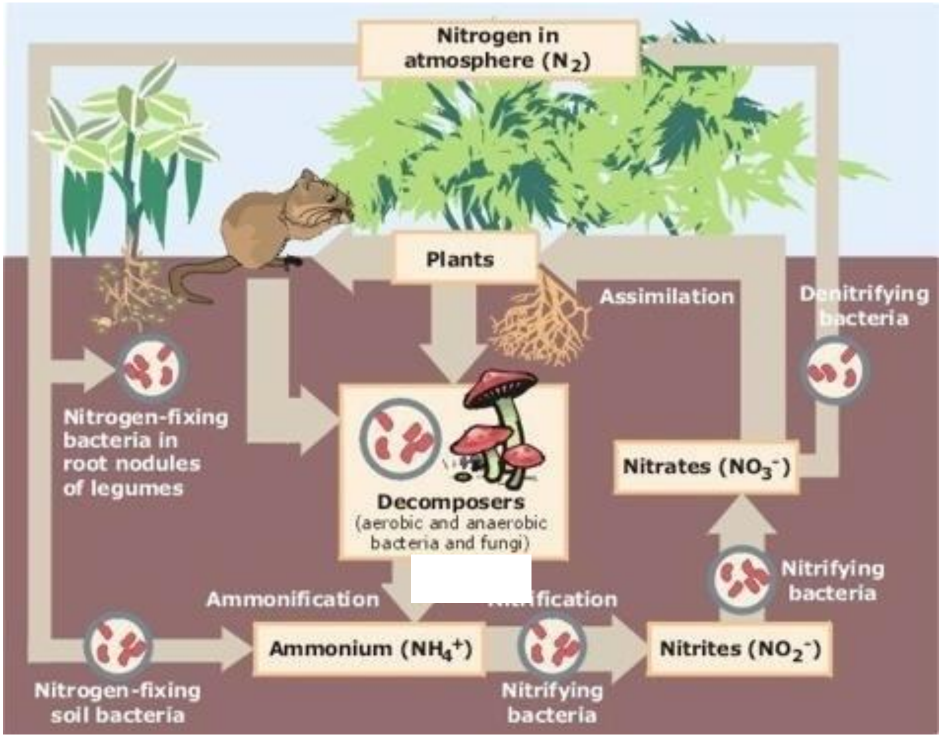

Fig. 2: Nitrogen Cycle

Source: http://www.epa.gov/maia/

3.4 Sulphur Cycle

Sulphur can exist in several oxidation states within organic and inorganic compounds. Oxidation-Reduction reactions mediated by microorganisms change the oxidation state of sulphur within various compounds establishing the sulphur cycle.

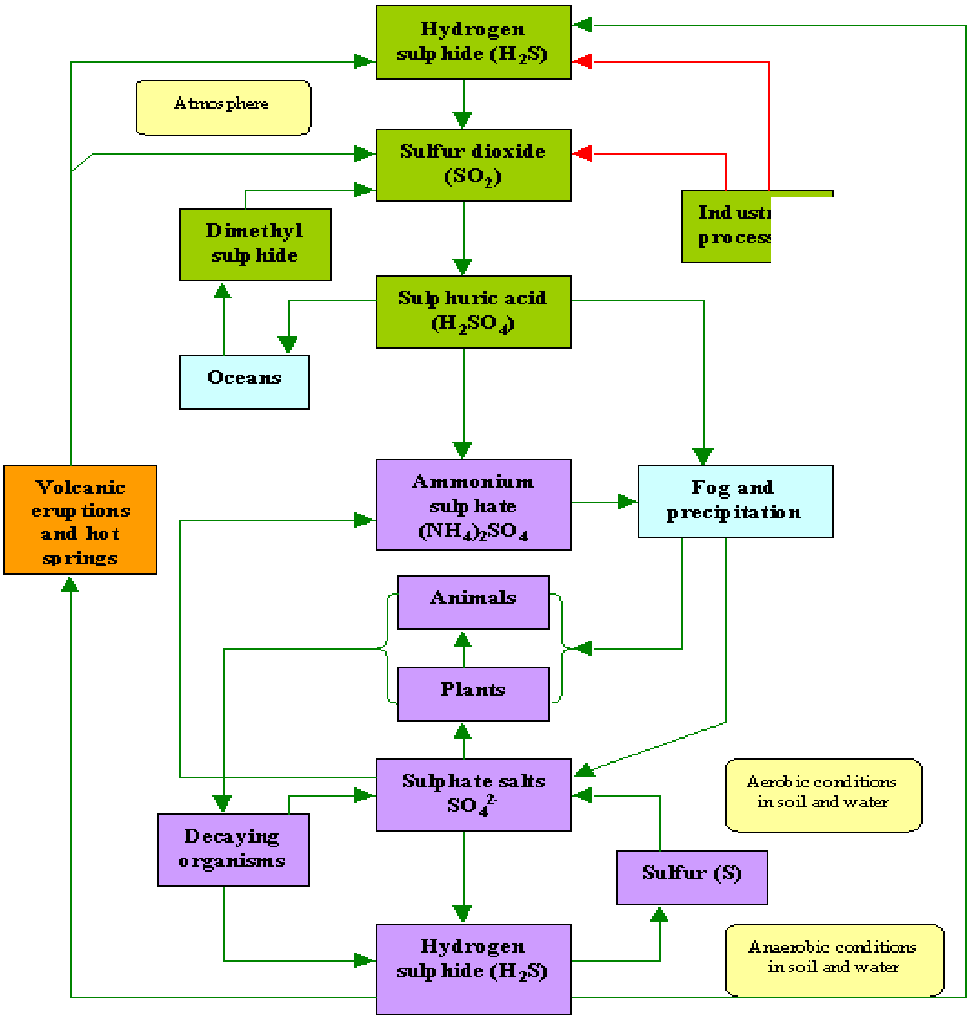

Fig. 3: Sulfur Cycle

Source: http://www.lenntech.com/images/sulfcycle.gif

Microorganisms are capable of removing sulphur from organic compounds under aerobic conditions, the removal of sulphur (desulfurization) of organic compounds results in the formation of sulphate, whereas under anaerobic conditions hydrogen sulphide is normally produced from the mineralization of organic sulphur compounds. Hydrogen sulphide may also be formed by sulphate-reducing bacteria that utilise sulphate as the terminal electron acceptor during anaerobic respiration. Hydrogen sulphide reacts with metals. Being negatively charged, it complexes or reacts easily with cations in the environment such as iron, aluminum and calcium. These compounds are relatively insoluble and most available to plants and microbes between pH 6 and 7 under these conditions. These organisms readily and rapidly convert phosphate to its organic form so that it becomes available to animals. The microbial transformation of phosphorus features the transformation of simple orthophosphate (PO_4^-) which bears phosphorus in the +5 valence state to more complex forms. These include the polyphosphates seen in metachromatic granules as well as more familiar macromolecules.

3.5 The Phosphorus Cycle

Biogeochemical cycling of phosphorus is important because all living cells require phosphorus for nucleic acids, lipids and some polysaccharides.

However, most environmental phosphorus is present in low concentration, locked within the earth's crust; hence it is the nutrient that limits growth. Unlike the Carbon and Nitrogen cycles, the phosphorus cycle has no gaseous component. Phosphorus is derived solely from the weathering of phosphate – containing rocks, hence in soil. Phosphorus exists in both inorganic and organic forms. Organic phosphorus is found in biomass, humus and other organic form. The phosphorus in these organic materials is recycled by microbial activity. Inorganic phosphorus, on the other hand is negatively charged, so it complexes or reacts easily with cations in the environment such as iron, aluminum and calcium. These compounds are relatively insoluble and most available to plants and microbes between pH 6 and 7.Under these conditions these organisms readily and rapidly convert phosphate to its organic form so that it becomes available to animals. The microbial transformation of phosphorus features the transformation of simple orthophosphate (PO_4^-) which bears phosphorus in the +5 valence state to more complex forms. These include the polyphosphates seen in metachromatic granules as well as more familiar macromolecules.

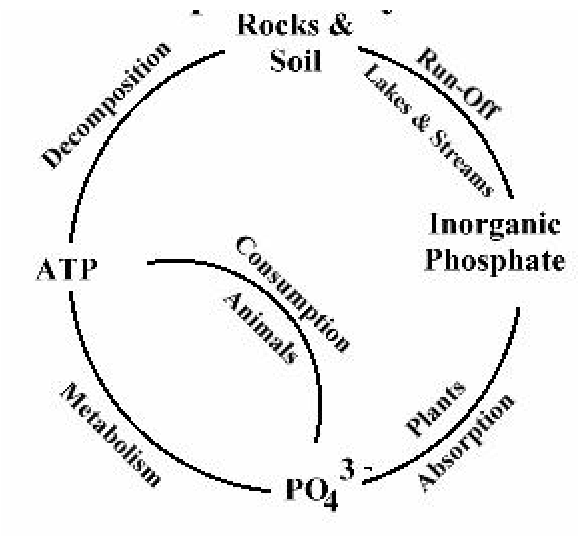

Fig. 4: Phosphorus Cycle

Source:
http://www.starsandseas.com/SAS%20Ecology/SAS%20chemcycles/cycle_phosphorus.htm

Self-Assessment Exercise

- Define the following terms
  i. Nitrogen Fixation
  ii. Nitrification
  iii. Denitrification
  iv. Ammonification

4.0 Conclusion

Microorganisms in the course of their growth and metabolism interact with each other to cycle nutrients such as carbon, sulphur and phosphorus. The nutrient cycling called biogeochemical cycling of elements involves both biological and chemical processes and is of global importance.

5.0 Summary

- Biogeochemical cycling of elements is the movement of materials via biochemical reactions through biospheres.
- It also refers to the biological and chemical processes that elements such as carbon nitrogen and sulfur undergo during microbial metabolism.
- Elements involved in biogeochemical cycles are used for three general purposes which are: (i) Biomass Production (ii) Energy Source and (iii) Terminal Electron Acceptor.
- The carbon cycle primarily involves the transfer of carbondioxide and organic carbon between the atmosphere where it occurs as principally as inorganic CO_2 and the hydrosphere and tithosphere which contain varying concentration of organic and inorganic compounds.
- The first step in carbon cycle is carbon fixation which is the conversion of CO_2 to organic matter by organisms such as *Cyanobacteria*.
- The second stage is the transfer of the fixed carbon from population to population within the biological community.
- The respiratory and fermentative metabolism of heterotrophic organisms returns inorganic carbon dioxide to the atmosphere.
- Decomposition of organic compounds such as plants and animals return CO2 to the cycle.
- Carbon is stored in rocks such as limestones ($CaCO_3$) and is dissolved as carbonate ions (CO_3) in oceans.
- Burning of fuels such as coal and petroleum releases carbon dioxide to the atmosphere.
- Nitrogen is an essential compound of DNA, RNA and proteins which are the building blocks of life.
- Atmospheric Nitrogen is not directly useable by most organisms but has to be converted to stable organic forms such as ammonium and nitrates.
- Three processes carried out by microorganisms in the nitrogen cycle are nitrogen fixation, nitrification and denitrification.
- Nitrogen fixation is strictly a bacterial process in which molecule nitrogen is converted to ammonium ion by a few bacteria that have a nitrogenase enzyme system.
- Nitrogen fixation is carried out by free-living nitrogen fixing bacteria such as *Azobacter, Cyanobacteria* and *Clostridium* found in the rhizosphere of plant.

- Symbiotic nitrogen fixing bacteria such as *Rhizobium* and *Bradyrhizobium* which in symbiotic association with leguminous plants help fix nitrogen in soil while *Cyanobacterium, Anabaenea* fix nitrogen when in association with the water fern.
- Nitrification is carried out by chemolithotropic bacteria which convert ammonium ions to nitrate (NO_3) ions in two steps.
- *Nitrosomonas* and *Nitrosospira* convert ammonia to nitrite while *Nitrococcus* and *Nitrobacter* convert nitrite to nitrate.
- Denitrification converts nitrate to dinitrogen.
- Ammonification is the decomposition process that converts organic nitrogen to ammonia (NH_3).
- Oxidation – Reduction reactions mediated by microorganisms change the oxidation states of sulphur with in various compounds establishing the sulphur cycle.
- Biogeochemical cycling of phosphorus is important because all living cells require phosphorus for nucleic acids, lipids and some polysaccharides.
- The phosphorus cycle has no gaseous state and phosphorus is derived solely from weathering phosphate containing rocks and organic phosphorus in biomass humus and other organic forms.

6.0 Self-Assessment Exercise

1. State three uses of biogeochemical cycles.
2. State specific examples of microorganisms.
3. Explain the role of symbiotic nitrogen fixing bacteria in biological nitrogen fixation in agricultural soils.
4. Outline the steps involved in nitrification.

7.0 References/Further Reading

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