MACHAKOS UNIVERSITY

CENTRE FOR OPEN, DISTANCE AND e-LEARNING

IN COLLABORATION WITH

SCHOOL OF PURE AND APPLIED SCIENCES

DEPARTMENT OF BIOLOGICAL SCIENCES

MODULE

SBT 418: MICROBIAL GENETICS

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MARCH 2024

COURSE OUTLINE

Purpose of the Course

The purpose of this course is to mechanisms behind stability and change in microbial genomes, information flow from DNA to proteins and the multiple levels at which gene expression can be regulated as well as the genetic aspects of extrachromosomal elements such as bacteriophages and plasmids.

COURSE OBJECTIVES

By the end of this course, the learner should be able to:

1. Understand gene expression control mechanisms
2. Describe importance of mutations and repair mechanism
3. Describe methods of DNA transfer
4. Demonstrate knowledge of the structure and function of plasmids and transposons

COURSE CONTENT

LECTURE 1: THE GENETIC MATERIAL

- Introduction to the genetic material and term definitions
- Structure of the genetic material
- Replication of DNA

LECTURE 2: PROTEIN SYNTHESIS

- Experimental proofs of DNA as the genetic material
- The central Dogma Theory

- The gene expression process

LECTURE 3: REGULATION OF GENE EXPRESSION

- Positive regulation; Inducible operon
- Negative regulation; repressible operon
- Comparison between prokaryotic and eukaryotic gene expression

LECTURE 4: MUTATIONS

- Definition of terms
- Types of mutations
- Causes of mutations

LECTURE 5: MUTATIONS

- Screening for successful mutants; negative and positive screening
- Effects of mutations; useful and harmful effects

LECTURE 6: DNA MEDIATED TRANSFORMATION

- The role of plasmids and transposons in transfer of genetic material
- Conjugation
- Griffiths classic experiment

LECTURE 7: DNA MEDIATED TRANSFORMATION

- Transduction
- Transformation

LECTURE 8: MICROBIAL QUORUM SENSING

- Process of biofilm formation
- Significance of biofilm formation

LECTURE 9: BACTERIOPHAGES

- bacteriophages structure
- bacteriophages life cycles and properties

LECTURE 10: DNA DAMAGE AND REPAIR MECHANISMS

- DNA damage
- Base excision
- Nucleotide excision
- mismatch repair mechanism.

COURSE REQUIREMENTS

Students are expected to:

1. Do proper semester registration.

2. Obtain Log-in credentials from ODEL Centre to enable them access the unit online.

3. Log in onto the online unit.

4. Actively participate in the online lecturers.

5. Actively participate in ALL e-tivities in each lecture.

6. Attempt self-test activities provided at the end of each lecture.

COURSE ASSESSMENT

i. There shall be Continuous Assessment Tests and End of semester Examination

ii. Continuous Assessment Tests (CATs) shall comprise 30%

iii. Continuous Assessment Tests shall comprise a computation to 30% of all marks garnered in

individual assignments, sit-in CATs, Practicals, group tasks and workshops which shall be assigned to you from time to time in the course of our online learning and interactions in the semester.

iv. End of semester Examination shall comprise 70%

v. End of semester Examination shall be a summative assessment of the content covered in

this unit.

LECTURE 1: THE GENETIC MATERIAL

1.1: INTRODUCTION

In this first lesson, we lay a foundation of the nature of the genetic material. It is paramount that the learner gets a brief history of the genetic material which entails a sequential order of experiments that were performed to ascertain the nature of the genetic material. Illustrations of the structure, building blocks of the genetic material and the bonding therein will demonstrate the actual nature of this material.

1.2: Lesson Objectives

By the end of the lesson, the learner should be able to:

1.2.1. Explain terms related to the genetic material

1.2.2. Illustrate the structure of DNA and RNA

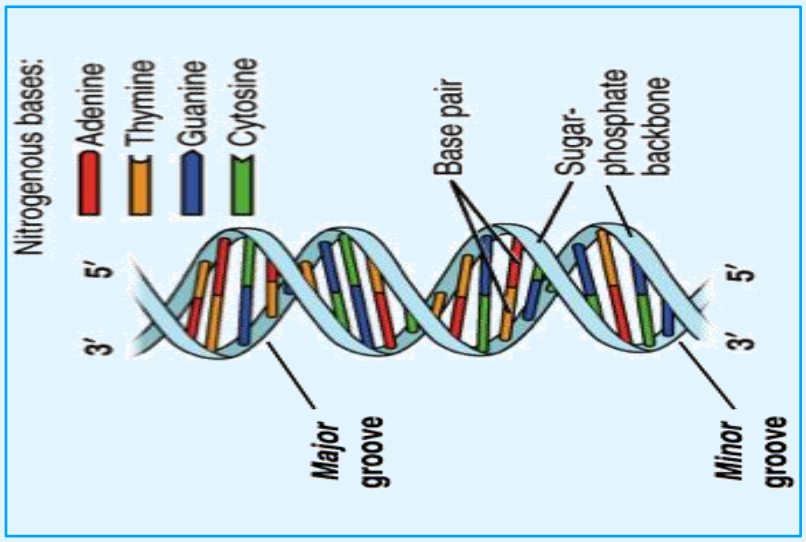
1.2.3. Describe the process of semi-conservative DNA replication

Structure of the genetic Material

More often we have come across terms such as deoxyribonucleic acids, ribonucleic acids, base pairs, double helix and many more. In this section we will understand and demystify these terms that seem complicated. We will also seek to understand the building blocks of DNA as well as illustrate how they bond to form the DNA double helix.

E-tivity 1

	1.2.2
Title	**Structure of the genetic Material**
Purpose	The purpose of this e-tivity is to illustrate the structure of DNA and the bonding therein.
Summary of overall task	Learners to watch video 1 a & b in the link below in order to grasp the structure of DNA https://drive.google.com/file/d/1Y6oEZZvrF36PUNo_HKmprwl6X948RrtT/view?usp=drive_link

	https://drive.google.com/file/d/1fnX9SH0E0vl9WMjdyWgo5LOhTKGX26bw/view?usp=drive_link
Spark	**ATTTTTTTGCCCCAATGCAAAAATGA,,,,What's the meaning of this sentence?** 
Individual task	a. Draw a DNA segment and illustrate the following: • Hydrogen bonding in complimentary bases • Phosphodiester bonding between adjacent nucleotides • Three and five prime ends b. Draw the structure of a Nucleotide and indicate the position of a pentose sugar, a phosphate group and a nitrogenous base.
Interaction begins	Leaners to describe the semi conservative process of DNA replication and give a justification for this process.
E-moderator interventions	1. Ensure that learners are focused on the contents and context of discussion. 2. Stimulate further learning and generation of new ideas. 3. Provide feedback on the learning progress. 4. Close the e-tivity
Schedule and time	This task should take one hour on 1st February 2024
Next	DNA and protein synthesis

Summary of the structure of Genetic Material

- DNA is a polymer—a large molecule that contains repeating units—composed of **2'-deoxyribose** (a five-carbon sugar), phosphoric acid, and the four nitrogen-containing bases denoted A, T, G, and C
- Two of the bases have a double-ring structure; these are called **purines.**
- The other two bases have a single-ring structure; these are called **pyrimidines.**
    - The purine bases are adenine (A) and guanine (G).
    - The pyrimidine bases are thymine (T) and cytosine (C).
- In DNA, each **base** is chemically linked to one molecule of the sugar deoxyribose and a phosphate group forming a compound called a **nucleotide**.
- Hence the nucleotide is the building block of a DNA molecule.
- In the absence of a phosphate group, the base is only attached to the deoxyribose sugar forming a nucleoside
- Thus a nucleoside is a nucleotide minus a phosphate.
- In the conventional numbering of the carbon atoms in the sugar in, the carbon atom to which the base is attached is the 1' carbon.

Summary of main features of semi conservative DNA replication

1. A progressive separation of the two strands of a DNA molecule.

2. Complementary base pairing of the bases located in the single stranded regions thus produced with the appropriate free deoxyribonulceotides.

3. Formation of phosphodiester linkages between the neighbouring deoxyribonucleotides that have base paired with the single stranded regions, thereby producing regions the new strand.

4. This ensures that the base sequence of the new strands are strictly complementary top those of the old strands.

5. The base sequence of a newly synthesized strand is dictated by the base sequence of the old strand, since the old strand serves as a template for the synthesis of the new strand.

LECTURE 2: PROTEIN SYNTHESIS

2.1: INTRODUCTION

The DNA sequence is a code that has to be decoded in order to produce the protein products for the cell's metabolism. Genes are sections of DNA that which when decoded give rise to specific polynucleotide sequences. The process of gene expression occurs in two steps: Transcription and Translation which have been summarized into the Central DOGMA theory.

2.2: Lesson Objectives

By the end of the lesson, the learner should be able to:

2.2.1. Explain terms related to the process of protein synthesis

2.2.2. Discuss in details the process of gene expression

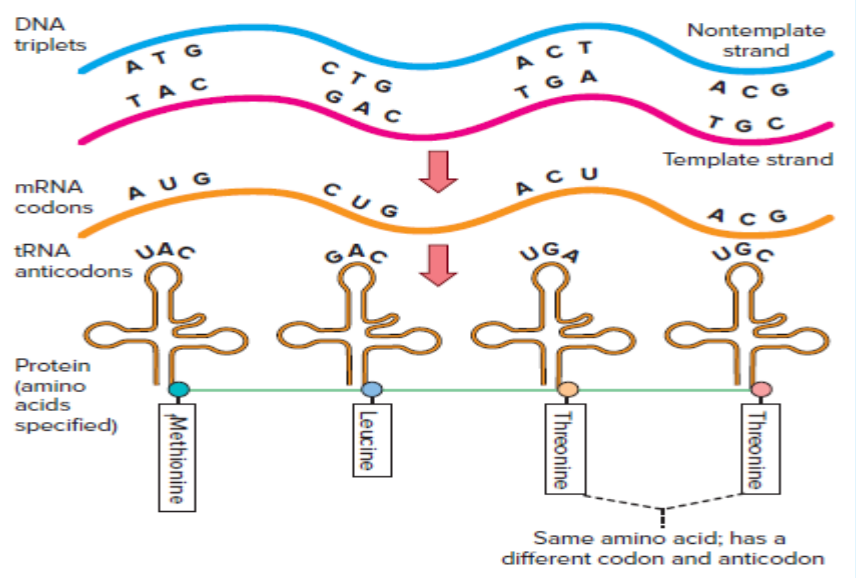
2.2.3. Describe the degenerate nature of the genetic code

Gene Expression

The master code of DNA is first used to synthesize RNA via a process called **transcription** and the information contained in the RNA is then used to produce proteins in a process known as **translation.** The language of DNA exists in the order of groups of three consecutive bases designated as **triplets** or **codons,** on one DNA strand**. O**ne gene differs from another in **the order** and **number of its triplets/codons**. An equally important part of this concept is that each triplet/codon represents a code for a particular amino acid. When the triplet code is transcribed and translated, it dictates the type and order of amino acids in a polypeptide (protein) chain as we will get to understand by the end of this lesson.

E-tivity 2

	2.2.2
Title	**Gene expression/Protein Synthesis**
Purpose	The purpose of this e-tivity is to illustrate and describe how DNA sequences (genes) are transcribed and then translated into proteins.
Summary of overall task	Learners to watch video 2 in the link below in order to grasp the processes of transcription and translation. https://drive.google.com/file/d/1025FVYCUcq4qAOROuNaqNtTzaj4RJbSa/view?usp=drive_link
Spark	**DNA** —**Transcription**→ **mRNA** —**Translation**→ **Protein**

	
Individual task	Describe the stages of transcription Describe the stages of translation
Interaction begins	Leaners to describe the gene expression process on LMS.
E-moderator interventions	1. Ensure that learners are focused on the contents and context of discussion. 2. Stimulate further learning and generation of new ideas. 3. Provide feedback on the learning progress. 4. Close the e-tivity
Schedule and time	This task should take one hour on 14th February 2024
Next	Regulation of Protein synthesis

Summary of Transcription and Translation

During **transcription**, an RNA molecule is synthesized using the codes on DNA as a guide or template. A large enzyme complex, **RNA polymerase,** is responsible for this process. This polymerase is more multipurpose than DNA polymerase, because the RNA enzyme works alone and does not require a helicase. Transcription proceeds in three stages: *initiation, elongation*, and *termination*. During **Initiation** the RNA polymerase recognizes a region on a gene called the **promoter region.** During elongation, the polymerase moves the transcription bubble forward, exposing subsequent sections of DNA. It simultaneously brings in nucleotides that are complementary to the DNA template and continues to assemble the mRNA strand in the 5′ to 3′

direction. **At termination**, the polymerase recognizes a site on DNA near the end of the gene that signals the separation and release of the completed mRNA, which will next enter the ribosome for translation. Translation also takes place in three stages: *Initiation, Elongation and Termination* which are completely different from those in transcription.

LECTURE 3: REGULATION OF GENE EXPRESSION

- Positive regulation; Inducible operon
- Negative regulation; repressible operon
- Comparison between prokaryotic and eukaryotic gene expression

3.1: INTRODUCTION

It is important for learners to understand why gene regulation cannot take place uncontrolled. The cell metabolic activities are controlled by enzymes and hormones which are products of gene expression. The production of these regulators of cell metabolism too must be controlled and the regulation of this production depends on factors that we seek to understand in this lesson.

3.2: Lesson Objectives

By the end of the lesson, the learner should be able to:

3.2.1 Distinguish between positive and negative regulation of gene expression

3.2.2 Illustrate specific examples of positive and negative gene regulation

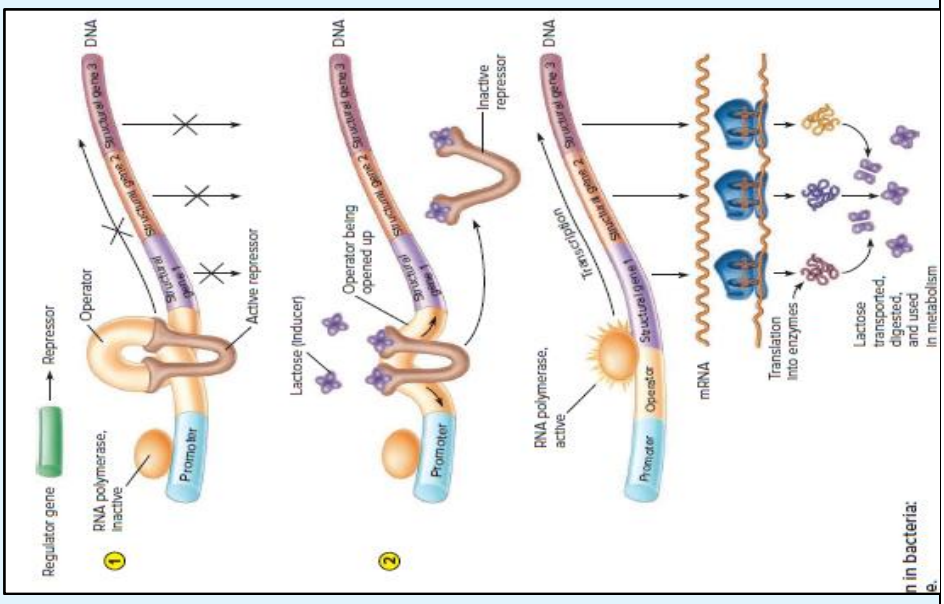
3.2.3 Differentiate prokaryotic and eukaryotic gene expression

Positive and Negative regulation of gene expression

In **positive regulation**, the product of gene expression is the protein enzyme required to break down a certain complex molecule to simple products. In a multistep degradative/catabolic system, the availability of the molecule to be degraded helps determine whether the enzymes in the pathway will be synthesized. In the presence of the degradable molecule, the enzymes of the degradative (catabolic) pathway are synthesized; in its **absence**, they are not. Such a system, in which the presence of a small molecule results in enzyme synthesis, is said to be **inducible** while the small molecule is called the **inducer.** A good example of a pathway with positive regulation is the Lac Operon and Lactose metabolism. **Negative Regulation** is found in the control of the synthesis of enzymes that participate in biosynthetic (anabolic) pathways; the final product of the pathway is frequently the regulatory

molecule. In the presence of the final product, the enzymes of the biosynthetic pathway are not synthesized; in its absence, they are synthesized. Such a system, in which the presence of a small molecule results in failure to synthesize enzymes, is said to be **repressible.** The excess product of the anabolic pathway that participates in the regulation is called the **co-repressor.** A good example of negative regulation of gene expression is the Arginine Synthesis Pathway.

E-tivity 3

	3.2.2
Title	**Regulation of Gene expression/Protein Synthesis**
Purpose	The purpose of this e-tivity is to help learners to understand how the positive and negative mechanisms of regulation of gene expression using specific examples.
Summary of overall task	Learners to watch video 3 in the link below in order to grasp the regulation mechanisms of gene expression. https://drive.google.com/file/d/1k-TA7r6cpYcsnJcEL0Ya4meXfpBiHfSa/view?usp=drive_link
Spark **(The Lac Operon)**	
Individual task	Describe the Lac operon Describe the Arginine synthesis operon

Interaction begins	Leaners to describe and compare gene expression in eukaryotes and prokaryotes.
E-moderator interventions	1. Ensure that learners are focused on the contents and context of discussion. 2. Stimulate further learning and generation of new ideas. 3. Provide feedback on the learning progress. 4. Close the e-tivity
Schedule and time	This task should take one hour on 21st February 2024
Next	Mutations

Summary of Gene expression Regulation

The **lactose (*lac*) operon** system, accounts for the regulation of lactose metabolism in *Escherichia coli.* It has three important features**:**

1. the **regulator,** composed of the gene that codes for a protein capable of repressing the operon (a **repressor**, **2. the *control locus***, composed of two areas, the promoter recognized by RNA polymerase and the operator and 3. **the *structural locus***, made up of three genes, each coding for a different enzyme needed to catabolize lactose. The enzymes β-galactosidase, hydrolyzes the lactose into its monosaccharides; permease, brings lactose across the cell membrane. In inducible system, *lac* operon is normally in an *off mode* and does not initiate enzyme synthesis when the appropriate substrate is absent. The key is in the repressor protein that is encoded by the regulatory gene. A substrate binding to one active site can distort a different active site and prevent it from accepting its substrate. In the absence of lactose, the repressor protein interacts with the operator and causes the operator to distort into a temporary loop configuration. This loop blocks access of the RNA polymerase to the DNA of the operator and prevents transcription. If lactose is added to the cell's environment, it triggers several events that turn the operon *on.* First, the binding of lactose to the repressor protein creates a shape change in the repressor that dislodges it from the operator segment**.** This opens up the operator to allow RNA polymerase to bind to it and begin transcription. The structural genes are transcribed.

LECTURE 4: MUTATIONS

- Definition of terms
- Types of mutations
- Causes of mutations

4.1: INTRODUCTION

Observable changes in living organisms which can be passed on from generation to generation are said to be heritable and they have a genetic basis. This lesson will bring to the knowledge of the learners the events that take place at the level of the genetic material so as to bring about phenotypic changes in organisms. These changes in the genetic material are also classifiable on the basis of their magnitude or the cause.

4.2: Lesson Objectives

By the end of the lesson, the learner should be able to:

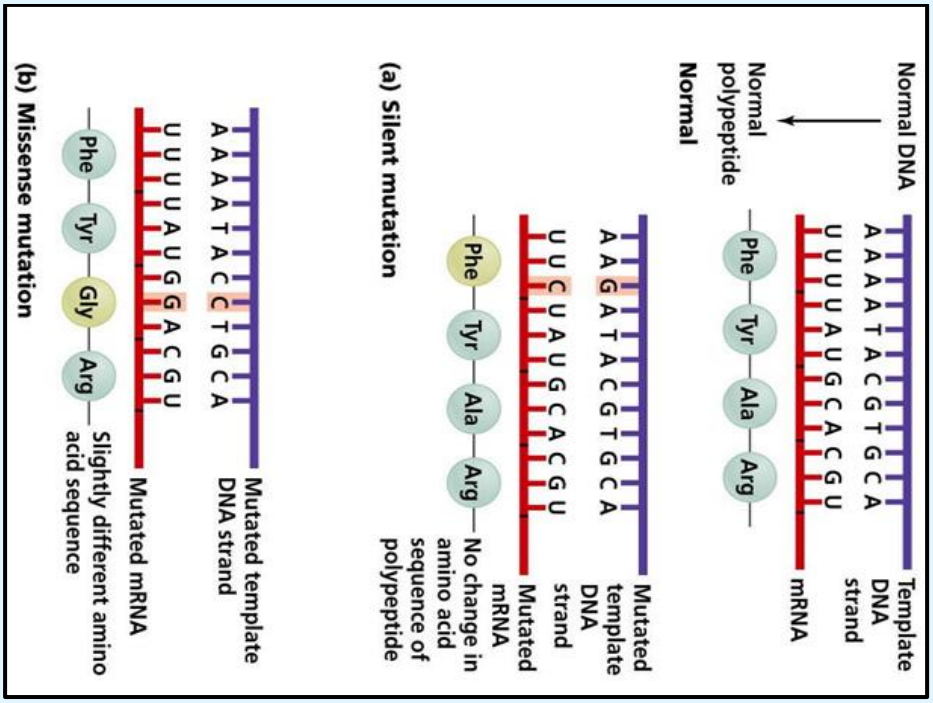
4.2.1. Define terms related to mutation process

4.2.2. Differentiate the various types of mutations

4.2.3. Describe the different causes of mutations

Mutation is one of the driving forces of evolution through addition of variation to populations of organisms. **A mutation** is a change/alteration in the DNA sequence of an organism. The mutation may become evident in altered gene expression, such as in the appearance or disappearance of anatomical or physiological traits. **Mutagens** are agents/substance that can bring about a permanent alteration to the physical composition of a DNA gene such that the genetic message is changed. Once the gene has been damaged or changed the mRNA transcribed from that gene will now carry an altered message. It can involve the loss of base pairs, the addition of base pairs, or rearrangement in the order of base pairs. A microorganism that exhibits a natural, non-mutated characteristic is known as a **wild type,** or wild strain. If a microorganism develops a mutation, it is called a **mutant strain.** Mutant strains can show variance in morphology, nutritional characteristics, genetic mechanisms, resistance to chemicals, temperature preference, and nearly any type of enzymatic function. Mutations are classified based on the **causative agent or the magnitude** of the genotypic change.

E-tivity 4

	4.2.2
Title	**Mutations: Causes and Types**
Purpose	The purpose of this e-tivity is to help learners to understand the genetic basis of mutations, types and causes of mutations.
Summary of overall task	Learners to watch video 4 in the link below in order to grasp the different types of mutations and their causes. https://drive.google.com/file/d/1OTajt9Z95kGZR2QvtT9c5h9Il9CX6M0A/view?usp=drive_link
Spark **(Point Mutations)**	
Individual task	Illustrate 4 types of point mutations Distinguish the different types of chromosomal mutations
Interaction begins	Learners to discuss the positive and detrimental effects of mutations with case studies.

E-moderator interventions	1. Ensure that learners are focused on the contents and context of discussion. 2. Stimulate further learning and generation of new ideas. 3. Provide feedback on the learning progress. 4. Close the e-tivity
Schedule and time	This task should take one hour on 28th February 2024
Next	Mutations

Summary of Causes of Induced Mutations

Induced mutations re caused by any agent that damages DNA, alters its chemistry, or in some way interferes with its functioning. These agents are called mutagens. Mutagens can be conveniently classified according to their mode of action. Three common types of chemical mutagens are:

A. Chemical Mutagens – They change the sequence of bases in a gene in a number of ways:

Mimic the correct nucleotide bases in a DNA molecule, but fail to base pair correctly during DNA replication. Remove parts of the nucleotide (such as the amino group on adenine), again causing improper base pairing during DNA replication. Add hydrocarbon groups to various nucleotides, also causing incorrect base pairing during DNA replication. E. g.

a. Base analogues

Are structurally similar to normal nitrogenous bases and can be incorporated into the growing polynucleotide chain during replication. Once in place, these compounds typically exhibit base-pairing properties different from the natural bases e.g.

- 5-bromouracil, an analog of thymine forms hydrogen bonds like cytosine, pairing with guanine rather than adenine. The mechanism of action of other base analogues is similar to that of 5-bromouracil.
- Nitrous Acid: Nitrous Acid affects DNA complementation. The acid randomly modifies the base adenine so that it will pair with cytosine instead of thymine. This change is made evident during DNA replication when a new base pair appears in daughter cells in a later generation.
- methyl-nitrosoguanidine adds methyl groups to guanine, causing it to mispair with thymine
- Hydroxylamine attaches a hydroxyl group (i.e., hydroxylates) to cytosine causing it to base pair like thymine.

b. Intercalating agents distort DNA to induce single nucleotide pair insertions and deletions.

They insert themselves (intercalate) between the stacked bases of the helix. This results in a mutation, possibly through the formation of a loop in DNA. Many mutagens, and indeed many carcinogens, damage bases so severely that hydrogen bonding between base pairs is impaired or prevented and the damaged DNA can no longer act as a template for replication.

B. Physical mutagens

For instance, ultraviolet (UV) radiation often generates thymine dimers between adjacent thymines. Exposure to direct UV light induces covalent linking between adjacent thymine nucleotides on a DNA strand forming a thymine dimer. These dimers cause the strand to buckle, disrupting normal base pairing. This prevents proper replication and transcription. Bacteria have enzymes to fix the damage created by UV light. An enzyme cuts the DNA at two point and removes the damaged portion. DNA polymerase synthesizes a new DNA segment using the healthly strand as a template. DNA ligase joins the new fragment to the old strand.

LECTURE 5: MUTATIONS

- Screening for successful mutants; negative and positive screening
- Effects of mutations; useful and harmful effects

5.1: INTRODUCTION

Experiments in which mutations are induced operate on a probability basis. As to whether a cell will pick up a mutation is trial and error. It is therefore paramount for researchers to set up experiments that will bring out the characteristics of the mutants against those of the wild type strains. These experiments are based on the knowledge of the effects the mutagen was intended to have on the organism and how they are expressed.

5.2: Lesson Objectives

By the end of the lesson, the learner should be able to:

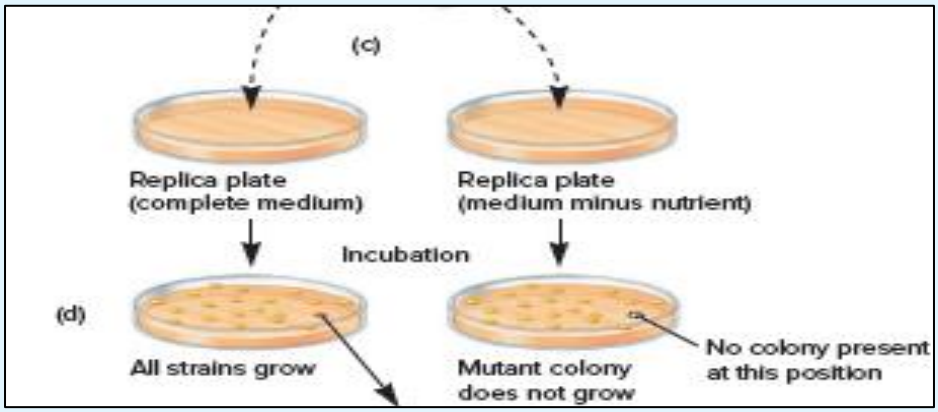
5.2.1 Define terms related to mutation process

5.2.2 Differentiate the various types of mutations

5.2.3 Describe the different causes of mutations

Positive selection of mutants involves experiments in which those organisms expressing altered traits upon mutation are identified and isolated while negative selection involves identifying colonies of microbes with ability to grow on incomplete media as the wild type. It goes further to identify regions of no growth on incomplete media and map the same on complete media as the mutant cells.

E-tivity 5

	4.2.2
Title	**Mutations: Screening for Mutants**
Purpose	The purpose of this e-tivity is to help learners design and describe experiments that are used to screen for mutant cells.
Summary of overall task	Learners to watch video 5 in the link below in order to grasp the regulation mechanisms of gene expression. https://drive.google.com/file/d/1XMPh9e9wHO3e7yz5C5MNxuAnrSAd_CFx/view?usp=drive_link
Spark (Negative Mutant selection)	
Individual task	Describe with an illustration the Replica Plate Method of mutant selection
Interaction begins	Learners to discuss the Ames test for Salmonella selection.
E-moderator interventions	1. Ensure that learners are focused on the contents and context of discussion. 2. Stimulate further learning and generation of new ideas. 3. Provide feedback on the learning progress. 4. Close the e-tivity
Schedule and time	This task should take one hour on 6th March 2024
Next	Transfer of Genetic Material

Summary of the Replica Plating Technique

This method is based on a variation between wild strains that can synthesize a nutrient, such as an amino acid, and mutant strains that cannot synthesize it. First a culture of test microbes is exposed to a chemical agent known to cause mutations and then plated on a complete medium that contains the particular nutrient. Colony position on the plate is retained by pressing a pickup or replica carrier onto the colonies and then using the carrier to **inoculate two plates in the same orientation**. These two plates contain: a complete medium containing the nutrient being tested for and incomplete medium that lacks it.
Colonies that grow on the complete medium could be of either the wild or mutant type, **but only the wild strains will grow on the incomplete medium**. By comparing the two plates side by side, it is possible to locate the position of the colony on the complete medium that does not grow on the incomplete one. This mutant colony is now detectable and can be isolated. Replica plating has been an effective technique to screen for phenotypic variants of bacteria and other microbes. It has the potential to provide information on a wide variety of metabolic characteristics. For example, it is used to detect mutants that vary in biochemical characteristics such as amino acid synthesis, substrate utilization, and sensitivity to antibiotics.

LECTURE 6: DNA MEDIATED TRANSFORMATION

6.1: INTRODUCTION

Naturally, bacteria exchange and acquire new genetic material in different ways. In this lesson we are going to explore the use of plasmids and transposons to transfer genetic material between cells. We will also endeavor to understand how bacterial cells in close contact can exchange genetic material using plasmids.

6.2: Lesson Objectives

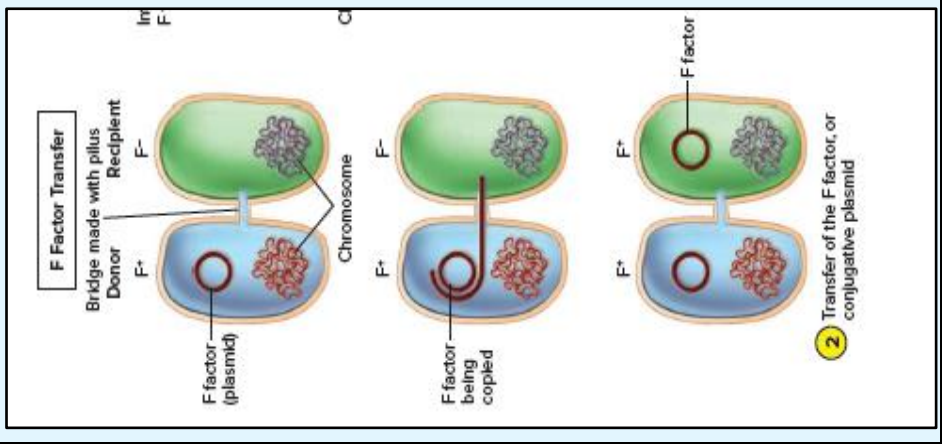
By the end of the lesson, the learner should be able to:

6.2.1 Explain the role of plasmids and transposons in the transfer of genetic material

6.2.2 Describe the mechanism of conjugation

Transmission of Genetic Material between bacterial cells typically involves small pieces of DNA in the form of **plasmids or chromosomal fragments.** While a plasmid can be stably replicated and inherited, chromosomal fragments must integrate themselves into the bacterial chromosome in order to be replicated and eventually passed to progeny cells. Depending upon the mode of transmission, the means of genetic recombination in bacteria can be **conjugation, transformation or transduction.** Conjugation requires the attachment of two cells and the formation of a bridge that can transport DNA.

E-tivity 6

	6.2.2
Title	**DNA Mediated Transformation: Conjugation**
Purpose	The purpose of this e-tivity is to help learners understand the role of plasmids in conjugation and the basis of Griffiths conjugation experiment.
Summary of overall task	Learners to watch video 6 in the link below in detailing the process of conjugation in bacteria. https://drive.google.com/file/d/1rYj58qzRnQaIDxHpS5mN5A5B-1Fwv0vJ/view?usp=drive_link
Spark (Conjugation)	

Individual task	Describe with an illustration the role of the F factor in bacterial conjugation
Interaction begins	Learners to discuss the significance of conjugation.
E-moderator interventions	1. Ensure that learners are focused on the contents and context of discussion. 2. Stimulate further learning and generation of new ideas. 3. Provide feedback on the learning progress. 4. Close the e-tivity
Schedule and time	This task should take one hour on 13th March 2024
Next	Transfer of Genetic Material: Transduction and Transformation

Summary of Conjugation

Conjugation is a mode of genetic recombination in which a plasmid or fragment of DNA is transferred from a donor cell to a recipient cell via a direct connection. Both gram-negative and gram-positive bacteria can conjugate, but only gram-negative cells operate with a specialized plasmid called a **fertility,** or **F factor.** This plasmid directs the synthesis of a unique **pilus,** also called a sex pilus, that functions in most conjugation transfers. The recipient cell is usually a related bacterium with a recognition site on its surface for interacting with the pilus. A cell's role in conjugation is denoted by F+ for the cell that has the F plasmid and by F− for the cell that lacks it. Contact is made when a pilus grows out from the F+ cell, attaches to the surface of the F cell, contracts, and draws the two cells together. At the site where the pilus attaches the two cells, a mating or conjugative bridge is formed that serves as a transfer system for the plasmid.

LECTURE 7: DNA MEDIATED TRANSFORMATION: Transduction & Transformation

7.1: INTRODUCTION

Viruses naturally infect some bacteria and such viruses are referred to as bacteriophages. When a phage that previously infected a bacterial host moves to a new host, it is likely to transfer genetic material between the two bacteria through the process of transduction. Also bacteria ae capable of

acquiring DNA from the surrounding aqueous environment through the process of transformation. In this lesson we will explore the two types of transduction and ways of making cells competent to acquire DNA from the surrounding solution.

7.2: Lesson Objectives

By the end of the lesson, the learner should be able to:

7.2.1 Distinguish generalized and special transduction processes

7.2.2 Describe the process of transformation and ways of making cells competent

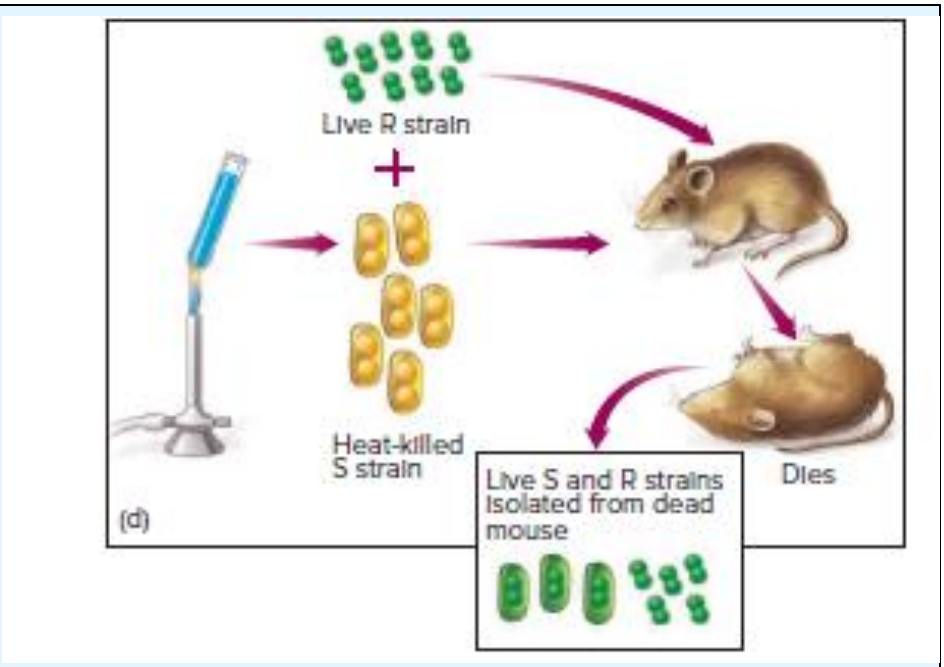
7.2.3 Describe Griffiths experiment

Bacteriophages (bacterial viruses) have been previously described as destructive bacterial parasites. Viruses can also serve as genetic vectors (an entity that can bring foreign DNA into a cell). The process by which a bacteriophage serves as the carrier of DNA from a donor cell to a recipient cell is **transduction.** Although it occurs naturally in a broad spectrum of bacteria, the participating bacteria in a single transduction event must be the same species because of the specificity of viruses for host cells.

Nonspecific acceptance by a bacterial cell of small fragments of soluble DNA from the surrounding environment is termed **transformation.** Transformation is facilitated by special DNA-binding proteins on the cell wall that capture DNA from the surrounding medium

E-tivity 7

	7.2.2
Title	**DNA Mediated Transformation: Transduction and Transformation**
Purpose	The purpose of this e-tivity is to help learners understand the two modes of transduction and the mechanism of transformation.
Summary of overall task	Learners to watch video 7 in the link below in understand the process of transduction in bacteria. https://drive.google.com/file/d/10x_MQyKBlv7N11lapjJflg-n48bZqJ-X/view?usp=drive_link

Spark (Transformation-Griffith's Experiment)	
Individual task	Describe with an illustration the role the two types of Transduction
Interaction begins	Learners to discuss the Griffiths conjugation experiment and ways of making cells competent.
E-moderator interventions	1. Ensure that learners are focused on the contents and context of discussion. 2. Stimulate further learning and generation of new ideas. 3. Provide feedback on the learning progress. 4. Close the e-tivity
Schedule and time	This task should take one hour on 20th March 2024
Next	Cell Conversation: Quorum Sensing

Summary of Specialized Transduction

A highly specific part of the host genome is regularly incorporated into the virus. This specificity is explained by the prior existence of a temperate prophage inserted in a fixed site on the bacterial chromosome. When activated, the prophage DNA separates from the bacterial chromosome, carrying a small segment of host genes with it. During a lytic cycle, these specific viral-host gene combinations are incorporated into the viral particles and carried to another bacterial cell.

Instances of transduction include the transfer of drug resistance seen in *Staphylococcus* spp. and the transmission of gene regulators in gram-negative rods such as *Escherichia* and *Salmonella*.

LECTURE 8: MICROBIAL QUORUM SENSING

- Process of biofilm formation
- Significance of biofilm formation

8.1: INTRODUCTION

Just like human beings, microbes too communicate with each other for various reasons. Hence they come together and are able sense presence of each other through production of certain metabolites that glue them together and help them to survive better and adapt to adverse environments.

8.2: Lesson Objectives

By the end of the lesson, the learner should be able to:

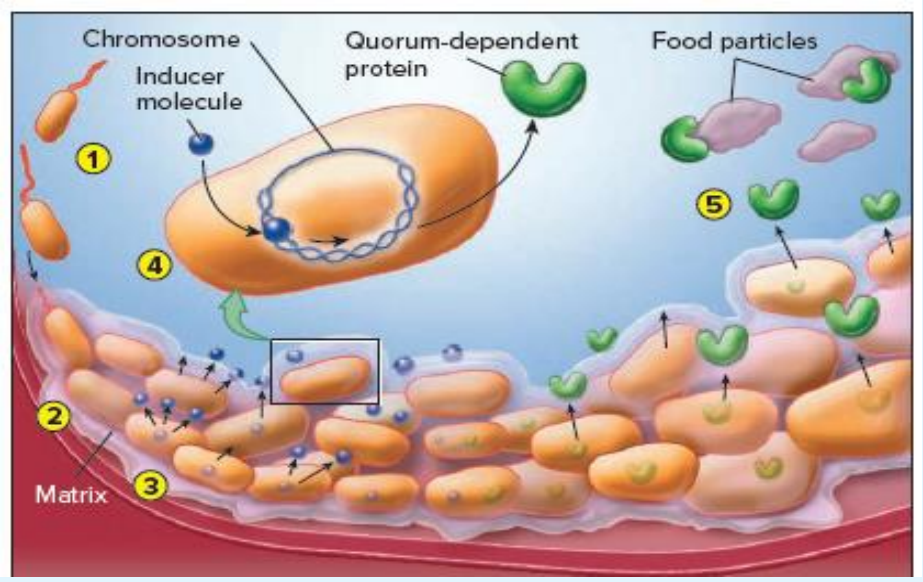
8.2.1 Describe the process of biofilm formation

8.2.2 Discuss the significance of biofilm in different environments

Biofilms result when organisms attach to a substrate by some form of extracellular matrix that binds them together in complex organized layers. Biofilms are so prevalent that they dominate the structure of most natural systems. **Quorum sensing** process occurs in several stages, including self-monitoring of cell density, secretion of chemical signals, and genetic activation**.**

E-tivity 8

	8.2.1
Title	**Microbial Communication: Quorum sensing**
Purpose	The purpose of this e-tivity is to help learners understand the process of biofilm formation and significance of biofilms.
Summary of overall task	Learners to watch video 8 in the link below in understand the process by which bacteria form biofilms. https://docs.google.com/presentation/d/0B2oR1SvfJoKzcTlodGhXc0FxTlU/edit?usp=drive_link&ouid=101967063945217221138&resourcekey=0-1G-14H611VTyFytK2XQt0g&rtpof=true&sd=true

Spark **(Biofilm Formation)**	
Individual task	Describe the mechanism of forming biofilms.
Interaction begins	Learners to discuss the significance of biofilms in natural systems.
E-moderator interventions	1. Ensure that learners are focused on the contents and context of discussion. 2. Stimulate further learning and generation of new ideas. 3. Provide feedback on the learning progress. 4. Close the e-tivity
Schedule and time	This task should take one hour on 27th March 2024
Next	Bacteriophages

Summary of the Biofilm Formation Process

A biofilm usually begins to form when a free-swimming (planktonic) bacterium attaches to a surface They more typically live in communities called biofilms. Cell-to-cell chemical communication, or quorum sensing, allows bacteria to coordinate their activity and group together into communities that provide benefits not unlike those of multicellular organisms. Therefore, biofilms are not just bacterial slime layers but biological systems; the bacteria are organized into a coordinated, functional community. Biofilms are usually attached to a surface, such as a rock in a pond, a human tooth or a mucous membrane. This community might be of a single species or of a diverse group of microorganisms. Biofilms also might take other, more varied forms. The floc that forms in certain types of sewage treatment is an example. In fast-flowing streams, the biofilm might be in the form of

filamentous streamers. Within a biofilm community, the bacteria are able to share nutrients and are sheltered from harmful factors in the environment, such as desiccation, antibiotics, and the body's immune system. The close proximity of microorganisms within a biofilm might also have the advantage of facilitating the transfer of genetic information by, for example, conjugation.

LECTURE 9: BACTERIOPHAGES

- bacteriophages structure
- bacteriophages life cycles and properties

9.1: INTRODUCTION

Viruses are obligate intracellular parasites of bacteria, protozoa, fungi, algae, plants, and animals which are ultramicroscopic size, ranging from 20 nm up to 450 nm (diameter). The basic structure consists of protein shell (capsid) surrounding nucleic acid core. The nucleic acid of the viral genome is either DNA or RNA but not both. The nucleic acid can be double-stranded DNA, single-stranded DNA, single-stranded RNA, or double-stranded RNA. Molecules on virus surface impart high specificity for attachment to host cell.

9.2: Lesson Objectives

By the end of the lesson, the learner should be able to:

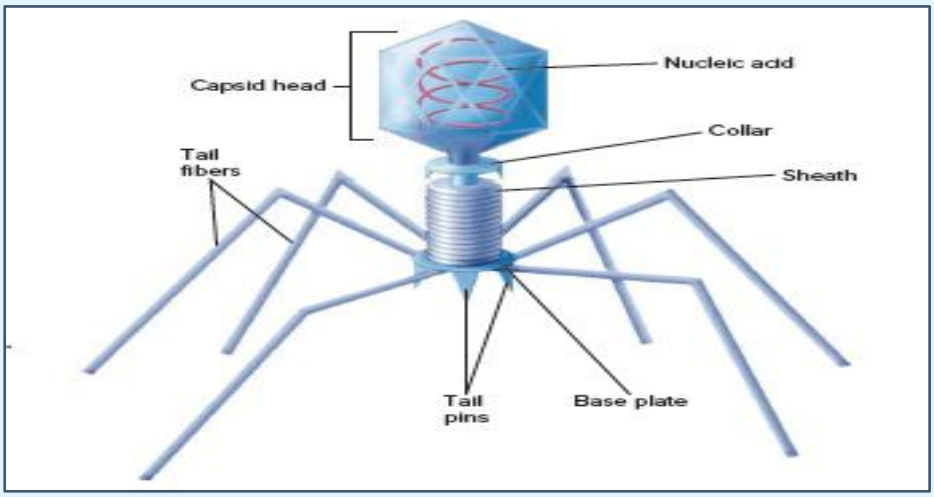
8.2.3 Describe the structure of bacteriophages

8.2.4 Describe the life cycle of viruses

The general phases in the life cycle of animal viruses are

1. Adsorption/attachment
2. Penetration and uncoating
3. Synthesis
4. Assembly
5. Release from the host cell

E-tivity 9

	9.2.1
Title	**Bacteriophages: Structure, Properties and life cycle**
Purpose	The purpose of this e-tivity is to expound on the structure of bacteriophages, their life cycle and properties.
Summary of overall task	Learners to watch video 9 in the link below to understand the structure and functioning of the bacteriophage structure. https://drive.google.com/file/d/1OB08TK0G8Fx05LkoQuHS8Swpa6EzRLta/view?usp=drive_link
Spark (T4 bacteriophage of *E. coli*)	
Individual task	Describe the stages of infection by bacteriophage
Interaction begins	Learners to distinguish the structure of the bacteriophage from animal viruses.
E-moderator interventions	1. Ensure that learners are focused on the contents and context of discussion. 2. Stimulate further learning and generation of new ideas. 3. Provide feedback on the learning progress. 4. Close the e-tivity
Schedule and time	This task should take one hour on 3rd April 2024
Next	DNA repair mechanisms

Summary of the stages of bacteriophage Infection

1 Attachment/adsorptionof the viral capsid (naked viruses) or of envelope components (enveloped viruses) to cell surface molecules (receptors). This involves specific interaction between viral glycoproteins (e.g. the haemagglutinin of influenza virus) and host cell surface components (e.g. N-acetylneuraminic acid for influenza virus). Many viruses have highly specific receptors, which limits the range of cell types that can be infected.

2 Penetration of the virus into the host cell (often by receptor-mediated endocytosis)

3 Uncoating follows, which involves the enzymatic removal of viral protein coat and liberation of nucleic acid and attached core proteins;

4 Production of virus-specific mRNA, in order to direct the host cell ribosome to produce viral proteins (core, capsid). The mechanisms for virus- specific mRNA production depend on the viral genome type: whether DNA or RNA, double or single stranded.

LECTURE 10: DNA DAMAGE AND REPAIR MECHANISMS

10.1: INTRODUCTION

DNA damage can happen due to normal cellular processes or exposure to damaging agents in the environment. The repair mechanisms of DNA repair can be divided into two general classes: direct reversal of the chemical reaction responsible for DNA damage, and (2) removal of the damaged bases followed by their replacement with newly synthesized DNA.

10.2: Lesson Objectives

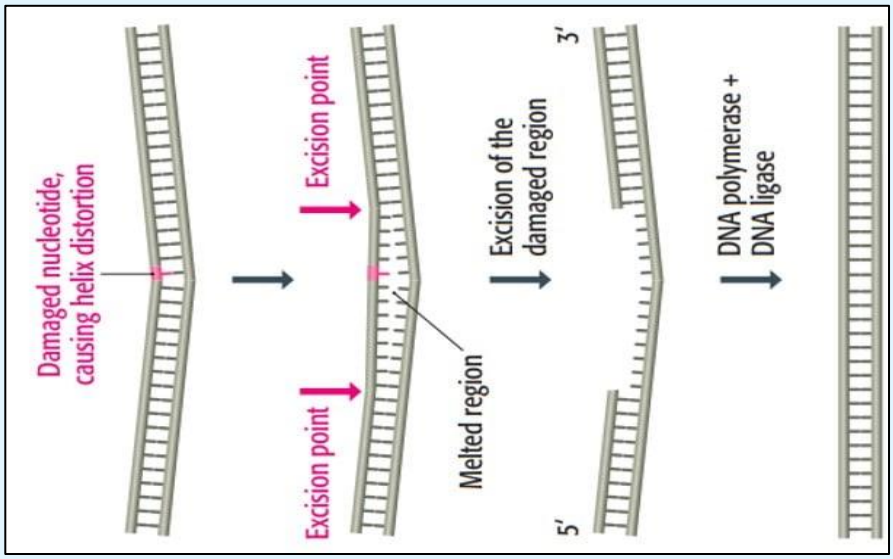
By the end of the lesson, the learner should be able to:

10.2.1 Describe the types and causes of DNA damages

10.2.2 Describe the types of repair mechanisms

DNA damage can also be classified into two types based on its origin or sources: endogenous and exogenous. Endogenous DNA damage originates from internal reactions involving chemically active DNA within cells such as Replication errors, Topoisomerase enzymes, Reactive oxygen species (ROS) and Alkylating agents. Exogenous DNA damage is caused by external factors, such as environmental agents, physical forces, or chemicals.

E-tivity 10

	9.2.1
Title	**DNA damage and Repair Mechanisms**
Purpose	The purpose of this e-tivity is help learners to understand various causes of DNA damage and repair mechanisms for each.
Summary of overall task	Learners to watch video 10 in the link below to help them describe the mechanisms the cell uses to repair damaged DNA. https://drive.google.com/file/d/1knoRrqvynFkv0UqHGW-72lBA0jzGOWJN/view?usp=drive_link
Spark **(Nucleotide excision repair)**	
Individual task	Describe Nucleotide excision repair, Direct reversal repair, Base excision repair and Mismatch repair
Interaction begins	Learners to discuss and illustrate the different causes of DNA damage.

E-moderator interventions	1. Ensure that learners are focused on the contents and context of discussion. 2. Stimulate further learning and generation of new ideas. 3. Provide feedback on the learning progress. 4. Close the e-tivity
Schedule and time	This task should take one hour on 10th April 2024
Next	REVISION, CATS and EXAM

Summary of the types of DNA damage

1. Endogenous DNA Damage

Endogenous DNA damage originates from internal reactions involving chemically active DNA within cells.

- **Replication errors** are one source of endogenous DNA damage that occurs during DNA replication when incorrect nucleotides are inserted opposite the template bases. During replication, some DNA polymerases with lower fidelity can be involved, leading to potential errors.
- **Topoisomerase enzymes** are another source of endogenous DNA damage. Topoisomerases remove the supercoiling of DNA during replication and transcription. However, misalignment of the DNA ends can stabilize the topoisomerase-DNA cleavage complex and result in the formation of DNA lesions.
- **Reactive oxygen species (ROS)** are produced during cellular processes and can cause oxidative damage to DNA. While ROS plays an important role in normal cellular functions, excessive levels can lead to various DNA lesions and modifications. Excessive ROS has been associated with the development of several human diseases like cancer, Alzheimer's disease, and diabetes.
- **Alkylating agents** are reactive compounds that can add methyl or ethyl groups to DNA bases, leading to chemical modifications. Spontaneous methylation events can generate different methylated bases. Some methylated bases are mutagenic and can lead to specific types of mutations.

2. Exogenous DNA Damage

Exogenous DNA damage is caused by external factors, such as environmental agents, physical forces, or chemicals.

- **Ionizing radiation (IR)** directly damages DNA or indirectly affects it through the generation of highly reactive hydroxyl radicals (•OH) from water molecules. IR can cause

different types of damage to the DNA such as base lesions, and single-strand and double-strand breaks.

- **Ultraviolet (UV) radiation** is another agent of DNA damage. It is the leading cause of skin cancers. UV light can form pyrimidine dimers where two pyrimidines on the same DNA strand are joined together. This alteration in DNA structure can block transcription and replication processes.
- **Exogenous alkylating agents**, found in sources like tobacco smoke and industrial activities, react with DNA and can cause mutagenic and carcinogenic changes. They primarily target the nitrogenous bases in DNA. Examples of alkylating agents include sulfur and nitrogen mustards.
- **Aromatic amines**, found in cigarette smoke, fuel, coal, dyes, and pesticides, are also exogenous sources of DNA damage. These agents can create long-lasting lesions in the DNA structure that lead to the substitution of DNA bases and frameshift mutations.
- **Polycyclic aromatic hydrocarbons (PAHs)** are known carcinogens found in sources like tobacco smoke, automobile exhaust, and other environmental pollutants. PAHs require activation by the liver's P-450 system to produce reactive substances that can potentially damage DNA.

Course Text Books

1. Microbial Genetics (Jones and Bartlett Series in Biology) 2nd Edition, Stanley Maloy (Author), ISBN-13: 978-0867202489
2. Genetics of Microbes, **Bainbridge** and Brian W. ISBN 978-1-4615-7093-6
3. Molecular Genetics of Bacteria 5th Edition Jeremy W. Dale (Author) and Simon F. Park (Author), ISBN-13: 978-0470741849

Course Journals

1. Microbial Genetics and Genomics ISSN: 2073-4425
2. Molecular Genetics and Genomics ISSN: 1617-4615
3. Genes & Genomics ISSN: 1976-9571

Reference Text Books

1. Molecular Genetics of Bacteria, 4th Edition 4th Edition Larry Snyder, Joseph E. Peters, Tina M. Henkin and Wendy Champness, ISBN-13: 978-1555816278
2. Modern Microbial Genetics, Uldis N Streips and Ronald E Yasbin, ISBN-13: 9780471386650
3. Clinical Microbiology Made Ridiculously Simple 6th **Mark Gladwin, Bill Trattler and C. Scott Mahan,** Edition ISBN-13: 978-1935660156.

Reference Journals

1. Microbial genetics ISSN: 2471-9315
2. Advances in Microbiology ISSN Print: 2165-3402
3. Medical Microbiology and Immunology ISSN: 0300-8584