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Course ………………………....Molecular Biology

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Dep ………………………. BS (MLT4th)

**Q1: Fill in the Blanks.**

1. The three main steps of PCR are Denaturation, Annealing and Extension.
2. The word “vaccine” originates from the Latin word Vaccinae.
3. Yeast is the oldest microbes exploited by humans for their benefit.
4. Restriction endonucleases are also called as Molecular Scissors.
5. Restriction map is a diagram or map of DNA molecule of an organism that shows specific sites of cleavage restriction sites.
6. A forensic technique used to identify individuals based on the variations in their DNAsequences is known as DNA finger printing.
7. Restriction modification system is mainly composed of Restriction Endonuclease and Methylase Enzyme

**Q2: Write short notes on the following**

1. Vaccines and its types
2. Biotechnology and its scope

 **1) Vaccines**

A vaccine is a substance that helps protect against certain diseases. Vaccines contain a dead or weakened version of a microbe. It helps your immune system recognize and destroy the living microbe during a future infection.

When inactivated or weakened disease-causing microorganisms enter the body, they initiate an immune response.

 This response mimics the body’s natural response to infection.

 These antigens trigger the production of “antibodies” by the immune system.

 Antibodies bind to corresponding antigens and induce their destruction by other immune cell.

**Types of vaccine**

There are four types of vaccine

1) Live

2) Dead

3) Subunit

4) Gentically engineered

 2) Biotechnology

Biotechnology is a technology in which we use biological system of whole living organisms or a part of organisms to create different products.

Scope of biotechnology

 B. Tech Biotech students have a vast scope as they can work in sectors of pharmaceuticals, agriculture, healthcare, food processing, nutrition, animal science, and environmental conservation. Not in terms of opportunities, even salary packages of biotechnology professionals are lucrative.

**Q3: Explain in detail the Restriction modification system.**

**Restriction-modification systems**

 Restriction-modification (R-M) systems are important components of prokaryotic defense mechanisms against invading genomes.

They occur in a wide variety of unicellular organisms, including bacteria and Achaea

They comprise two contrasting enzymatic activities:

 -Restriction endonuclease (REase)

 -Methyltransferase (MTase).

Phage (or viruses) invade all types of cells.

Bacteria are one favorite target.

Defense mechanisms have been developed by bacteria to defend themselves from these invasions.

The system they possess for this defense is the restriction-modification system.

This system is composed of a

* **Restriction endonuclease**
* **Methylase enzyme**

**E**ach bacterial species and strain has their own combination of restriction and methylating enzymes

Restriction enzyme - an enzyme that cuts DNA at internal phosphodiester bonds; different types exist and the most useful ones for molecular biology (Type II) are those which cleave at a specific DNA sequence

Methylase - an enzyme that adds a methyl group to a molecule; in restriction-modification systems of bacteria a methyl group is added to DNA at a specific site to protect the site from restriction endonuclease cleavage

**Q4: What are Different types of retraction enzymes? Recombinant DNA, Recombinant DNA technology and its application.**

There are four main types of retraction enzymes.

* Type I
* Type II
* Type III
* Type IV

 Traditionally, four types of restriction enzymes are recognized, designated I, II, III, and IV, which differ primarily in structure, cleavage site, specificity, and cofactors. Types I and III enzymes are similar in that both restriction and methylase activities are carried out by one large enzyme complex, in difference to the type II system, in which the restriction enzyme is self-determining of its methylase. Type II restriction enzymes also change from types I and III in that they cut DNA at specific sites within the respect site; the others cleave DNA randomly, sometimes hundreds of bases from the recognition sequence. Numerous thousand type II restriction enzymes have been recognized from a variety of bacterial species. These enzymes make out a few hundred distinct sequences, usually four to eight bases in length. Type IV restriction enzymes cleave only methylated DNA and show weak sequence specificity.

 **Recombinant DNA**

Recombinant DNA technology is a technique in which we combine different constituent of organisms to form DNA.

**Recombinant DNA Technology**

Using Recombinant DNA technology, we can isolate and clone single copy of a gene or a DNA segment into an indefinite number of copies, all identical.

Simply defined, it is the art of cutting and pasting genes**.**

**Applications**

* Restriction enzyme site analysis
* Polymerase chain reaction (PCR)
* Genetic engineering
* Recombinant DNA libraries
* DNA sequencing
* Mutation studies
* Transformation

**Q5: As students of MLT how will you use Restriction endonuclease in lab?**

**Use Restriction endonuclease in lab**

In the lab we will use restriction enzyme digestion to estimate the similarity in nucleotide sequence between different DNA molecules. This can be accomplished by comparing the DNA fragments that end result from digestion of each molecule. Let’s accept that you have a molecule of DNA that is 10,000 bases (10 kilobases, or 10kb) in length that you are working with. If that molecule has the sequence GAATTC 4kb from one end then digestion with Eco RI will generate two fragments: one that is 4kb in length and one that is 6kb in length. Using gel electrophoresis next digestion you will be able to imagine the result. Any DNA molecule with the same sequence will always yield these two fragments when digested by Eco RI but DNA molecules with not the same sequences will be cut into fragments of different sizes. Therefore, the basic idea behind this technique is quite simple. To compare two different DNA samples you digest them with the same restriction enzyme and use gel electrophoresis to examine the fragments generated. If the fragments are different then the two samples contain DNA molecules with different nucleotide sequences. Notice, however, that the opposite is not necessarily true, if the two examples yield the same fragments they may still have different sequences. Therefore, we repeat this for a number of different restriction enzymes. If the samples continuously yield the same fragments then they are either very similar or identical in nucleotide sequence