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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 **variolae vaccinae** **(cowpox).**
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 DNA sequences 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

Answer.

1. Vaccine:

The word "vaccine" is derived from a Latin variolae vaccinae (cowpox) which Edward Jenner demonstrated in 1798 could prevent smallpox in humans.
Today the term "vaccine" applies to all biological preparation, produced from living organisms, that enhance immunity against disease and either prevent (prophylactic vaccines) or, in some cases treat disease (therapeutic vaccines).

Types of vaccines

i. Live
ii. Dead
iii. Subunit

i. Live vaccine:

These vaccines are composed of live, attenuated microorganisms that caused a limited infection in their host sufficient to induce an immune response, but insufficient to caused disease.

ii. Dead vaccine:

When it is unsafe to use live microorganisms to prepare vaccine they are dead or inactivated.
These are preparation of the normal (wild type) infectious microorganisms that have been rendered nonpathogenic, usually by treatment with using heat so that they cannot replicate at all.

iii. Subunit vaccine:

The subunit vaccine is a fragment of a pathogen, typically a surface protein, that is use to trigger an immune response and stimulate acquired immunity against the pathogen from which it is derived.

2. Biotechnology:

Biotechnology is the manipulation of living organism and organic material to serve human needs.

- The science of using living organism or the products of living organisms for the benefit of humans and their surroundings.

Examples:

- Yeast and bread making and alcohol production
- Use of beneficial bacteria to kill harmful organisms
- Cloning of plants and animals
- Artificial insemination

Scope of Biotechnology:

There has been increased activity and research between different agricultural areas with common research techniques and goals.

Plant Science:

Improvement of varieties according to relevant agronomic features:

- **Productivity** (resistance to biotic stress: pests, viruses, pathogens, abiotic stress tolerance to drought, salinity ... herbicide tolerance. Plant-soil interaction, nutrient absorption, metabolism improvement, etc.)
- **Nutrition improvement:** vitamin enrichment, flavor enhancement.

Animal Science

- Increased use of methods of in vitro fertilization and artificial insemination improve selected breed programs
- Transgenic (also known as recombinant DNA) is the transferal of a specific gene from one organism to another.
- To date, cattle, sheep, pigs, goats, horses, mules, cats, rats and mice have been cloned, beginning with the first cloned animal, a sheep named Dolly, in 1996.

Environmental Science:

- Use of biotechnology techniques in environmental science for cleaning contaminants and protecting endangered species
- Bioremediation-use of natural organisms to clean contaminants
- Immunoassay tests are used to test for the presence of contaminants in soil, water and even blood
- Installation of biological barriers to prevent the transfer of harmful microorganisms between production facilities

Example: Tire wash channels

Health/Agri-medicine:

- Pharming-the creation of plants and animals capable of producing medical substances
- The use of biological barriers to prevent the spread of harmful microorganisms that could contaminate food sources
- DNA analysis/paternity testing has emerged as a technique to test the genetic ancestry of animals.

Q3: Explain in detail the Restriction modification system.

Answer:

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 archaea
- 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-modificiation 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.

Answer:

Types of restriction enzymes;

Types	Cleavage site	Location of methylase	Examples
Type i	Random around 1000bp away from recognition site	Endonuclease and methylase location on a single protein molecule	Ecok I EcoA I CfrAI
Type ii	Specific within the recognition site	Endonuclease and methylase are separate entities	Ecop I BamH I Hind III
Type iii	Random 24-26 bp away from recognition site	Endonuclease and methylase lacation on a single protein molecule	EcoP I Hinf III EcoP 151

Recombinant DNA:

- DNA molecules formed by laboratory methods of genetic recombination (such as molecular cloning) to bring together genetic material from multiple sources
- This is DNA that has been formed artificially by combining constituents from different organisms.

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.

Application of rDNA Technology:

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

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

Answer:

In the laboratory, restriction enzymes (or restriction endonuclease) are used to cut DNA into smaller fragments. The cuts are always made at specific nucleotide sequences.

Different restriction enzymes recognize and cut different DNA sequences. Restriction enzymes can be isolated from bacterial cells and used in the laboratory to manipulate fragments of DNA, such as those that contain genes, for this reason they are indispensable tools of recombinant DNA technology (genetic engineering).

The end