

Important Questions

Multiple Choice Questions:

- 1. Rising of dough is due to
 - (a) Multiplication of yeast
 - (b) Production of CO₂
 - (c) Emulsification
 - (d) Hydrolysis of wheat flour starch into sugars.
- 2. An enzyme catalysing the removal of nucleotides from the ends of DNA is:
 - (a) endonuclease
 - (b) exonuclease
 - (c) DNAligase
 - (d) Hind-II.
- 3. The transfer of genetic material from one bacterium to another through the mediation of a vector-like virus is termed as:
 - (a) Transduction
 - (b) Conjugation
 - (c) Transformation
 - (d) Translation.
- 4. Which of the given statement is correct in the context of observing DNA separated by agarose gel electrophoresis?
 - (a) DNA can be seen in visible light
 - (b) DNA can be seen without staining in visible light
 - (c) Ethidium bromide stained DNA can be seen in visible light
 - (d) Ethidium bromide stained DNA can be seen under exposure to UV light.
- 5. 'Restriction' in Restriction enzyme refers to:
 - (a) Cleaving of phosphodiester bond in DNA by the enzyme
 - (b) Cutting of DNA at specific position only
 - (c) Prevention of the multiplication of bacteriophage in bacteria
 - (d) All of the above.
- 6. A recombinant DNA molecule can be produced in the absence of the following:
 - (a) Restriction endonuclease
 - (b) DNA ligase
 - (c) DNA fragments
 - (d) E. coli.
- 7. In agarose gel electrophoresis, DNA molecules are separated on the basis of their:

- (a) Charge only
- (b) Size only
- (c) Charge to size ratio
- (d) All of the above.
- 8. The most important feature in a plasmid to be used as a vector is:
 - (a) Origin of replication (ori)
 - (b) Presence of a selectable marker
 - (c) Presence of sites for restriction endonuclease
 - (d) Its size.
- 9. While isolating DNA from bacteria, which of the following enzymes is not used?
 - (a) Lyozyme
 - (b) Ribonuclease
 - (c) Deoxyribonuclease
 - (d) Protease.
- 10. Which of the following has popularised the PCR (polymerase chain reactions)?
 - (a) Easy availability of DNA template
 - (b) Availability of synthetic primers
 - (c) Availability of cheap deoxyribonucleotides
 - (d) Availability of 'Thermostable' DNA polymerase.
- 11. An antibiotic resistance gene in a vector usually helps in the selection of:
 - (a) Competent cells
 - (b) Transformed cells
 - (c) Recombinant cells
 - (d) None of the above.
- 12. Significance of 'heat shock' method in bacterial transformation is to facilitate:
 - (a) Binding of DNA to the cell wall
 - (b) Uptake of DNA through membrane transport proteins
 - (c) Uptake of DNA through transisent pores in the bacterial cell wall and plasma membrane
 - (d) Expression of antibiotic resistance gene.
- 13. A biotechnologist wanted to create a colony of E.coli possessing the plasmid pBR322, sensitive to Tetracycline. Which one of the following restriction sites would he use to ligate a foreign DNA?
 - (a) Sal I
 - (b) Pvu I

(c) EcoRI

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- (d) Hind III
- 14. Which of the following steps are catalysed by Taq polymerase in a PCR reaction?
 - (a) Denaturation of template DNA
 - (b) Annealing of primers to template DNA
 - (c) Extension of primer end on the template DNA
 - (d) All of the above.
- 15. A bacterial cell was transformed with a recombinant DNA that was generated using a human gene. However, the transformed cells did not produce the desired protein.

 Reasons could be:
 - (a) Human gene may have intron which bacteria cannot process
 - (b) Amino acid codon for humans and bacteria are different
 - (c) Human protein is formed but degraded by bacteria
 - (d) All of the above.

Very Short Question:

- 1. A restriction enzyme digests DNA into fragments. Name the technique used to check the progression of this enzyme and separate DNA fragments.
- 2. Name two commonly used vectors in genetic engineering.
- **3.** Some enzymes are considered as molecular scissors. in genetic engenrring. What is the name assigned to such enzymes?
- 4. Write conventional nomenclature of EcoRI.
- **5.** A linear DNA fragment and a plasmid has three restriction sites for EcoRlhow many fragments will be produced from linear DNA and plasmid respectively.
- **6.** An extra chromosomal segment of circular DNA of a bacterium is used to carry gene of interest into the host cell. What is the name given to it?
- **7.** Name the substance used as a medium in gel electrophoresis.
- 8. What is Bioconversion?
- **9.** Name the bacterium that yields thermostable DNA polymerase.
- 10. Which enzymes are known as "molecular Scissors"?

> Short Questions:

- 1. Name two main steps which are collectively referred to as down streaming process. Why is this process significant?
- 2. What are 'Selectable marker'? What is their use in genetic engineering?
- 3. How can the desired product formed after genetic engineering be produced on a

commercial scale?

- **4.** What is "Insertional Inactivation"?
- 5. What are the two basic techniques involved in modern Biotechnology?
- 6. Differentiate between plasmid DNA and chromosomal DNA?
- 7. What is the role of enzyme "Ligase" in genetic Engineering?
- 8. Name the components a bioreactor must possess to achieve the desired product?

Long Questions:

- 1. How is recombinant DNA transferred to host?
- 2. Explain any three methods of vector less gene transfer.
- 3. Write a note on the cloning vector.

> Assertion and Reason Questions:

- 1. Two statements are given-one labelled Assertion and the other labelled Reason. Select the correct answer to these questions from the codes (a), (b), (c) and (d) as given below.
 - a) Both assertion and reason are true and reason is the correct explanation of assertion.
 - b) Both assertion and reason are true but reason is not the correct explanation of assertion.
 - c) Assertion is true but reason is false.
 - d) Both assertion and reason are false.

Assertion: The insertion of DNA fragment into pBR 322 plasmid using enzyme Pst I or Pvu I make ampicillin resistant gene non functional.

Reason: Bacterial cells containing recombinant pBR322 is unable to grow in the presence of ampicillin.

- 2. Two statements are given-one labelled Assertion and the other labelled Reason. Select the correct answer to these questions from the codes (a), (b), (c) and (d) as given below.
- a) Both assertion and reason are true and reason is the correct explanation of assertion.
- b) Both assertion and reason are true but reason is not the correct explanation of assertion.
- c) Assertion is true but reason is false.
- d) Both assertion and reason are false.

Assertion: Soil inhabiting bacterium Agrobacterium tumefaciens is called a natural plant genetic engineer.

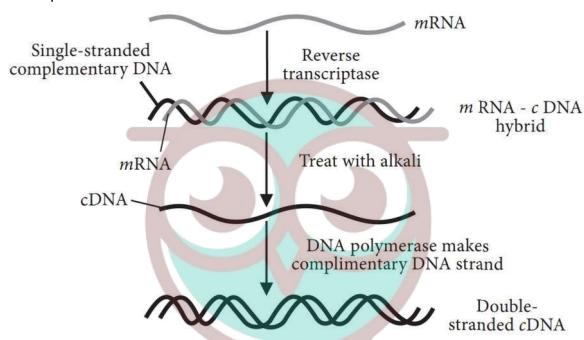
Reason: Agrobacterium tumefaciens produce crown galls in several dicot plants.

Case Study Questions:

1. Read the following and answer any four questions from (i) to (v) given below:

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The DNA, which is transferred from one organism into another by joining it with the vehicle DNA is called passenger or foreign DNA. Generally three types of passenger DNAs are used. These are complementary DNA (cDNA), synthetic DNA (sDNA) and random DNA. Complementary DNA (cDNA) is synthesized on RNA template (usually mRNA) with the help of reverse transcriptase. Synthetic DNA (sDNA) is synthesized on DNA template or without a template. Random DNA are small fragments fanned by breaking a chromosome of an organism in the presence of restriction endonucleases.



- (i) Reverse transcriptase enzyme was discovered by:
 - a) Temin and Baltimore.
 - b) Cohen and Boyer.
 - c) Arber and Nathan.
 - d) Paul Berg.
- (ii) During cDNA formation, what would happen if DNA formed by reverse transcriptase is not treated with the alkali?
 - a) cDNA will not be digested.
 - b) mRNA will not be digested.
 - c) Hydrogen bonds will not form between base pairs.
 - d) rnRNA will not be formed.
- (iii) Enzyme that helps in the formation of double stranded cDNA is:
 - a) DNA synthetase
 - b) Ligase
 - c) DNA polymerase

- d) Helicase.
- (iv) DNA polymerase can be obtained form:
 - a) Retrovirus.
 - b) Agrobacteriurn.
 - c) Tobacco mosaic virus.
 - d) Thermus aquaticus.
 - (v) DNA synthesised without a template is referred to as:
 - a) Complementary DNA.
 - b) Random DNA.
 - c) Synthetic DNA.
 - d) Z-DNA..
 - 2. Read the following and answer any four questions from 9(i) to 9(v) given below:

Bioreactors are considered as vessels in which raw materials are biologically converted into specific products by microbes, plant and animal cells or their enzymes. They are used for large scale production as they provide optimum growth conditions such as temperature, pH, substrate, vitamins, oxygen and salts for obtaining desired product. Most commonly used bioreactors are of stirring type which include simple stirred tank bioreactor and sparged stirred-tank bioreactor.

- (i) Bioreactor are useful in:
 - a. Amplifying a gene.
 - b. Isolation of genetic material.
 - c. Processing large volume of culture.
 - d. Infecting DNA in a cell.
- (ii) Which of the following is essential to obtain desired product in a bioreactor?
 - a. Size of the bioreactor.
 - b. Sterile condition.
 - c. Quantity of the raw material.
 - d. All of these.
- (iii) Assertion: The stirred-tank is well suited for large scale production of microorganisms under aseptic conditions.

Reason: In sparged stirred tank bioreactor, surface area for oxygen transfer is increased.

a. Both assertion and reason are true and reason is the correct explanation of assertion.

- b. Both assertion and reason are true but reason is not the correct explanation of assertion.
- c. Assertion is true but reason is false.
- d. Both assertion and reason are false.
- (iv) Growth condition that could affect the quality of obtained product in a bioreactor are:
 - a. Temperature and pH only.
 - b. pH and oxygen supply only.
 - c. Temperature and oxygen supply only.
 - d. Temperature, pH and oxygen supply.
- (v) Vessels in which raw materials are biologically converted into specific products are.
 - a. Bioreactors.
 - b. Fermentors.
 - c. Gene guns.
 - d. Both (a) and (b).

✓ Answer Key-

Multiple Choice Answers:

- 1. (b) Production of CO₂
- 2. (b) exonuclease
- 3. (a) Transduction
- 4. (d) Ethidium bromide stained DNA can be seen under exposure to UV light.
- **5.** (d) All of the above.
- **6.** (d) E. coli.
- **7.** (b) Size only
- 8. (a) Origin of replication (ori)
- **9.** (c) Deoxyribonuclease
- **10.** (d) Availability of 'Thermostable' DNA polymerase.
- **11.** (b) Transformed cells
- **12.** (c) Uptake of DNA through transisent pores in the bacterial cell wall and plasma membrane
- **13.** (a) Sal I
- 14. (c) Extension of primer end on the template DNA

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15. (a) Human gene may have intron which bacteria cannot process

Very Short Answers:

- 1. Gel electrophoresis
- 2. Plasmid and Bacteriophage.
- 3. Restriction Enzymes.
- **4.** E. = Escherichia; co = coli; R = Name of Strain; I = order in which enzyme isolated from strain of bacteria.
- Number of fragments of linear DNA = 4Number of fragments of plasmid = 3
- 6. Plasmid.
- 7. Agarose
- **8.** Bioconversion refers to the process by which raw material are converted to specific product by microbial, plant or animal cell.
- 9. Thermusaquaticus.
- 10. Restriction Endonuclease.

> Short Answer:

1. Separation and Purification

This process is essential because before reaching into market, the product has to be subjected for clinical trial and quality control.

- **2.** A selectable marker is a gene which helps in selecting those host cells which contains the vector & eliminating the non–transformanteg gene encoding resistance to antibiotics are usefulSelectable markers as they allow Selective growth of transformants only.
- **3.** The product obtained from genetic engineering is subjected to a series of processes collectively called downstream processing before it made into final processes involved in downstreamprocessing are :- Separation & purification.
- **4.** If a recombinant DNA is inserted within the coding Sequence of enzyme B–galactosidase. This results into inactivation of enzyme which is referred to as "Insertional Inactivation". The presence of chromogenic Substrate gives blue–coloured colonies if the plasmid in bacteria does not have an insert presence of insert results into insertional inactivation & the colonies do not produce anycolor.
- 5. The two basic techniques involved in modern Biotechnology are:
 - i. Genetic Engineering is the technique of altering the nature of genetic material or introduction of it into another host organism to change its phenotype.
 - ii. Techniques to facilitate the growth & multiplication of only the desired microbes or cells

in large number under sterile conditions for manufacture

- **6.** Plasmid DNA is extranuclear DNA, found in protoplasmic whereas chromosomal DNA is the nuclear or genetic DNA which is found within the nucleus.
- **7.** Enzyme "Ligase" acts as molecular Suture which helps in joining two pieces of DNA. The Joining process requires ATP as it derive energy to construct phosphodiester bond between cohesive ends.
- **8.** Enzyme "Ligase" acts as molecular Suture which helps in joining two pieces of DNA. The Joining process requires ATP as it derive energy to construct phosphodiester bond between cohesive ends.

Long Answer:

- 1. Transfer of recombinant DNA into the host:
 - i. The bacterial cells must be made competent to take up DNA; this is done by treating them with a specific concentration of calcium, that increases the efficiency with which DNA enters the cell through the pores in its cell wall.
 - ii. Recombinant DNA can then be forced into such cells by incubating the cells with recombinant DNA on ice followed by placing them at 42°C and then putting them back on ice (heat shock treatment),
 - iii. Microinjection is a method in which the recombinant DNA is directly injected into the nucleus of the animal cell with the help of microneedles or micropipettes.
 - iv. Gene gun or biolistics is a method suitable for plant cells, where cells are bombarded with high-velocity microparticles of gold or tungsten coated with DNA.
 - v. Disarmed pathogens are used as vectors; when they are allowed to infect the cell, they transfer the recombinant DNA into the host.
- 2. Vectors of gene transfer. Following are common methods of vectors gene transfer.
 - Microinjection: Microinjection is the process/technique of introducing foreign genes into a host cell by injecting the DNA directly into the nucleus by using microneedle or micropipette.
 - ii. Electroporation: Electroporation is the process by which transient holes are produced in the plasma membrane of the (host) cell to facilitate entry of foreign DNA.
 - iii. Gene Gun: Gene gun is the technique of bombarding microprojectiles (gold or tungsten particles) coated with foreign DNA with great velocity into the target cell.

3. Cloning vectors:

- i. Plasmids and bacteriophages are the commonly used vectors
- ii. Presently genetically engineered/ synthetic vectors are also used for easily linking the foreign DNA and selection of recombinants from non-recombinants.

- iii. The following features are required to facilitate cloning in a vector:
 - (a) Origin of replication (Ori)
 - (b) Selectable marker
 - (c) Cloning (Recognition) site
 - (d) Small size of the vector.

> Assertion and Reason Answers:

1. (b) Both assertion and reason are true but reason is not the correct explanation of assertion.

Explanation:

Plasmid pBR322 has a variety of unique restriction sites for restriction endonucleases. Two unique sites, Pst I and Pvu /are located within the amp' gene and Bamrll, Sal I etc. are within tet¹ gene. The presence of restriction sites within the marker tef and amp' permits an easy selection for cell transformed with the recombinant pBR322. Insertion of the DNA fragment into the plasmid using enzyme Pst I and Pvu I places the DNA insert within the gene amp' and make it non functional.

2. (b) Both assertion and reason are true but reason is not the correct explanation of assertion.

Case Study Answers:

1.

- (i) (a) Temin and Baltimore.
- (ii) (b) mRNA will not be digested.

Explanation:

The cDNA formation involves the alkaline denaturation of the mRNA-cDNA hybrid. The double stranded DNA molecule formed after the activity of reverse transcriptase is treated with alkali to digest mRNA.

In a bioreactor, all operations must be carried under sterile conditions to avoid contamination.

(iii) (c) DNA polymerase

Explanation:

A cDNA strand is formed on the separated single stranded DNA template with the help of DNA polymerase enzyme.

- (iv) (d) Thermus aquaticus.
- (v) (c) Synthetic DNA.

2.

(i) (c) Processing large volume of culture.

Explanation:

Bioreactors are considered as vessels in which raw molecules are biologically converted into specific products.

(ii) (b) Sterile condition.

Explanation:

In a bioreactor, all operations must be carried under sterile conditions to avoid contamination.

- (iii) (b) Both assertion and reason are true but reason is not the correct explanation of assertion.
- (iv) (d) Temperature, pH and oxygen supply.

Explanation:

A bioreactor provides the optimal growth conditions such as temperature,pH, substrate, vitamins, oxygen and salts for obtaining the desired product.

(v) (d) Both (a) and (b).

