BIOTECHNOLOGY – PRINCIPLES & PROCESS

1. Match the scientists in Column-I with their related discoveries in Column-II and select the correct option from the codes given below.

Column I	Column II	
A. Kary Mullis	(i)	Father of genetic engineering
B. Paul Berg	(ii)	Noel prize for the discovery of restriction endonulceases
C. Stanley Cohen and Hebert Boyer	(iii)	Developed polymerase chain reaction
D. Arber, Smith and Nathan	(iv)	Isolated an antibiotic resistant gene from a plasmid of the bacterium *Salmonella typhimurium*

(A) A-(iii), B-(i), C-(iv), D-(ii) (B) A-(iii), B-(iv), C-(i), D-(ii) (C) A-(iv), B-(ii), C-(iii), D-(i) (D) A-(i), B-(iii), C-(iv), D-(ii)

2. Plasmid used to construct the first recombinant DNA was isolated from which bacterium species?

(A) *Escherichia coli*

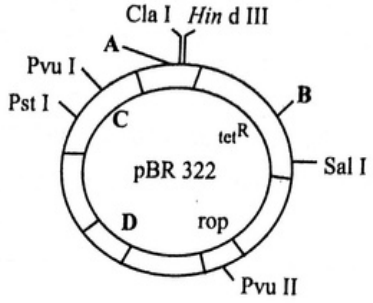
(B) *Salmonella typhimurium*

(C) *Agrobacterium tumefaciens*

(D) *Thermus aquaticus*

3. Which of the following are parts of biotechnology?
   (i) *In vitro* fertilization (ii) Synthesis of a gene (iii) Correcting a defective gene (iv) Developing a DNA vaccine (A) (i) and (ii) (B) (ii) and (iii) (C) (iii) and (iv) (D) (i), (ii), (iii) and (iv)

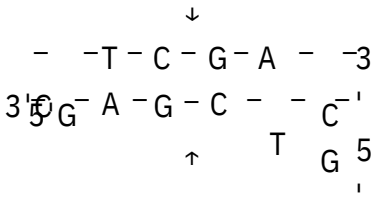
4. Identify A, B, C and D in the given figure of *E.coli* cloning vector pBR 322 and select the correct option.



	A	B	C	D
(A)	*Hid* d I	*Eco* R I	ampR	ori
(B)	*Hid* d I	*Bam* H I	kanR	ampR
(C)	*Bam*H I	Pst I	ori	ampR
(D)	*Eco* R I	*Bam* H I	ampR	ori

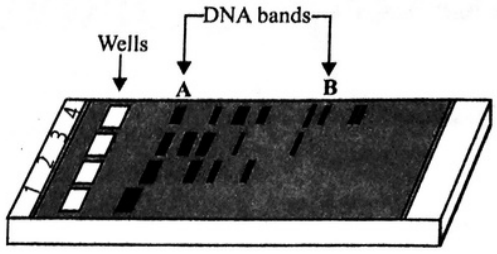
5. Read the following statements and select the correct ones.
   (i) Electrophoresis is a technique used for the separation of substances based on their size and change.
   (ii) Plasmids are extra-chromosomal, self-replicating, usually circular, double stranded DNA molecules found naturally in many bacteria and also in some yeast.
   (iii) It is not advisable to use an exonuclease enzyme while producing a recombinant DNA molecule.
   (iv) In *Eco* RI, the roman numeral I indicates that it was the first enzyme isolated from *E.coli* RY 13.
   (A) (i) and (ii)
   (B) (iii) and (iv)
   (C) (i), (ii) and (iv)
   (D) (i), (ii), (iii) and (iv)

6. The restriction enzyme responsible for the cleavage of following sequence is



   (A) *Eco* R I
   (B) *Hind* II
   (C) *Bamm* HI
   (D) *Eco* R II

7. Study the given figure carefully and select the incorrect statements regarding this.



(i) It represents a typical agarose gel electrophoresis in which are 1 contains undigested DNA.

(ii) Smallest DNA bands are formed at A and largest DNA bands are formed at B.

(iii) The separated DNA fragments can be visualized after staining in the visible light.

(iv) The separated DNA bands are cut out from the agarose gel and extracted from the gel piece. This step is known as elution.

(A) (i) and (ii) (B) (ii) and (iii)

(C) (ii) and (iv) (D) (i) and (iv)

8. Smallest unit of DNA capable of recombination.

(A) Genome (B) Plasmid (C) Recon (D)Vector

9. Which of the following statements is not correct regarding *Eco* R I restriction endonuclease enzyme?

(A) It is isolated from *Escherichia coli* RY 13

(B) Its recognition sequence is 5'–GAATTC–3'
3'–CTTAAG–5'

(C) It produces complimentary blunt ends

(D) None of these

10. Read the following statements and select the correct ones.

(i) Same kind of sticky ends are produced when a DNA has been cut by different restriction enzymes.

(ii) Exonucleases make cuts at specific positions within the DNA.

(iii) *Hin d* II was the first restriction endonuclease to be isolated.

(iv) A bacteriophage has the ability to replicate within bacterial cells by integrating its DNA with bacterial DNA.

(v) Presence of more than one recognition sites within the vector facilitates the gene cloning.

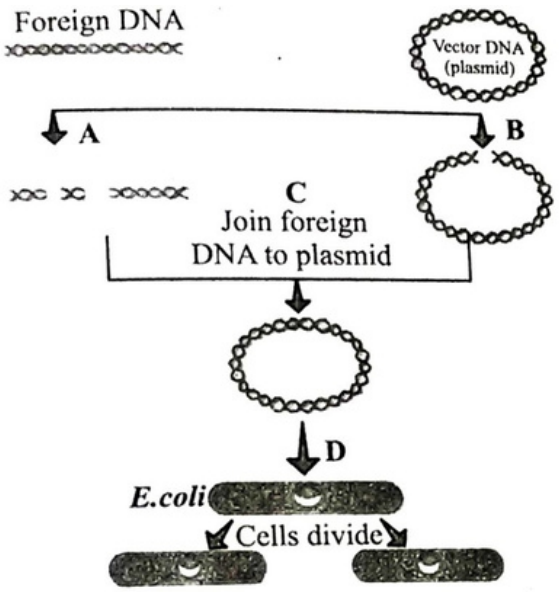
(A) (i), (iii) and (v)

(B) (i) and (iv)

(C) (iii) and (iv)

(D) (ii), (iii) and (iv)

11. The flow chart given below represents the process of recombinant DNA technology. Identify A, B, C and D.



(A) A-Restriction endonuclease, B-Restriction exonuclease, C-DNA ligase, D-Transformation

(B) A-Restriction endonuclease, B-Restriction endonuclease, C-DNA ligase, D-Transformation

(C) A-Restriction endonuclease, B-Restriction endonuclease, C-Hydrolase, D-Transformation

(D) A-Restriction endonuclease, B-Restriction endonuclease, C-Hydrolase, D-Transduction

12. Which of the following tools of recombinant DNA technology is incorrectly paired with its use?

(A) *Eco*RI – Production of sticky ends

(B) DNA ligase – Multiplication of *r*DNA molecules

(C) DNA polymerase – In polymerase chain reaction to amplify sections of DNA

(D) Selectable marker – Identification of transformants

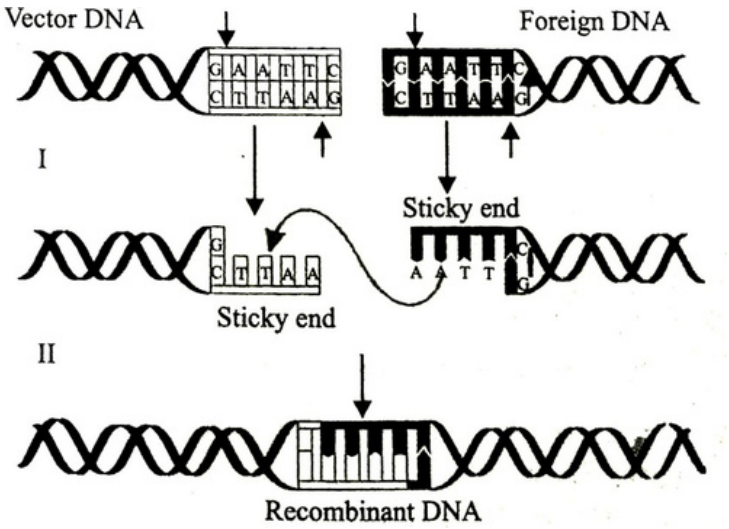
13. In recombinant DNA technology, the term vector refers to

(A) the enzyme that cuts DNA into restriction fragments
(B) the sticky end of a DNA fragment
(C) a plasmid used to transfer DNA into a living cell
(D) a DNA fragment which carries only *ori* gene.

14. The correct sequence of making a cell competent is

(A) treatment with divalent cations → incubation of cells with recombinant DNA on ice → heat shock (42°C) → placing on ice
(B) heat shock (42°C) → incubation of cells with recombinant DNA on ice → treatment with divalent cations → placing on ice
(C) treatment with divalent cations → placing on ice → incubation of cells with recombinant DNA on ice → heat shock (42°C)
(D) incumbation of cells with recombinant DNA on ice → heat shock (42°C) → treatment with divalent cations → placing on ice

15. Study the following figures and identify the enzymes involved in steps I and II.



(A) *Eco* R I and DNA Ligase
(B) *Hind* II and DNA Ligase
(C) *Eco* R I and *Hind* II
(D) Restriction endonuclease and exonuclease

16. Which of the following statements are correct?
(i) Restriction enzymes cut the strand of DNA a little away from the centre of the palindrome site, but between the same two bases on the opposite strands.
(ii) *Hind* II always cuts DNA molecules at a particular point by recognizing a specific sequence of six base pairs.
(iii) Separated DNA fragments cannot be visualized without staining on an agarose gel electrophoresis.
(iv) 'Ori' is the sequence responsible for controlling the copy number.
(v) DNA is positively charged molecule.
(A) (i), (iii) and (v)

(B) (i), (ii), (iii) and (iv)
(C) (iii), (iv) and (v)
(D) (i), (ii), (iii), (iv) and (v)

17. Select the correct option to fill up the blanks.
(i) _____ a natural polymer extracted form _____.
(ii) The DNA fragments purified by gel electrophoresis are used in constructing _____ by joining them with _____.
(iii) The ligation of alien DNA is carried out at a _____ present in one of the two _____ in a plasmid vector.
(iv) _____ remains active during the high temperature induced denaturation of ds DNA.
(v) DNA fragments are resolved according to their _____ through _____ in agarose gel electrophoresis.
(A) (i) Agarose, sea weeks (ii) recombinant DNA, cloning vector (iii) restriction site, antibiotic resistance genes (iv) *Taq* polymerase (v) size, sieving effect
(B) (i) Restriction site, antibiotic resistance genes (ii) agarose, sea weeds (iii) recombinant DNA, cloning vector (iv) *Taq* polymerase (v) size, sieving effect

(C) (i) Agarose, sea weeds (ii) restriction site, antibiotic resistance genes (iii) recombinant DNA, cloning vector (iv) *Taq* polymerase (v) size, sieving effect

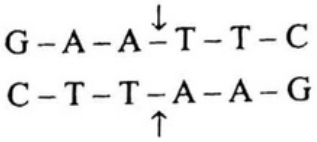
(D) (i) Size, sieving effect (ii) agarose, sea weeds (iii) recombinant DNA, cloning vector (iv) *Taq* polymerase (v) restriction site, antibiotic resistance genes

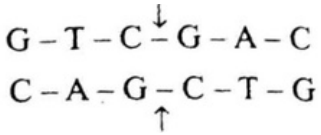
18. How many fragments will be generated if you digest a linear DNA molecule with a restriction enzyme having four recognition sites on the DNA?

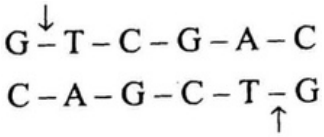
(A) 3 (B) 6

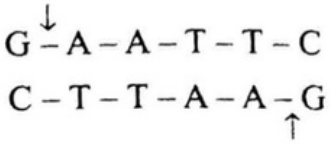
(C) 5 (D) 4

19. Which of the following correctly depicts the recognition site for *Eco* R I?

(A) 

(B) 

(C) 

(D) 

20. If a person obtains transformants by inserting a recombinant DNA within the coding sequence of enzyme β-galactosidase, he will separate out recombinants from non-recombinants by which of the following observations?

(A) Non-recombinant colonies do not produce any colour whereas recombinants give blue coloured colonies
(B) Recombinant colonies do not produce any colour whereas non-recombinants give blue coloured colonies
(C) Recombinants and non-recombinants both produce blue coloured colonies
(D) No colonies are formed due to insertional inactivation.

21. _____, a crown gall bacterium, is called as 'natural genetic engineer' of plants.
(A) *Escherichia coli*
(B) *Streptomyces albus*
(C) *Agrobacterium tumefaciens*
(D) *Azotobacter*

22. Which of the following sequences is recognized by restriction enzyme *Bam* H I?

(A) 5′ – GAATTC – 3′ / 3′ – CTTAAG – 5′

(B) 5′ – AAGCTA – 3′ / 3′ – TTCGAT – 5′

(C) 5′ – GGATCC – 3′ / 3′ – CCTAGG – 5′

(D) 5′ – CCCAAT – 3′ / 3′ – GGGTTA – 5′

23. Read the given statements and select the correct option.

Statement 1 : In insertional inactivation, blue colour produced by bacterial colonies indicates that the plasmid does not have an insert into the bacterial genome.

Statement 2: Presence of insert result into insertional inactivation of β-galactosidase enzyme and the colonies do not produce any colour.

(A) Both statements 1 and 2 are correct and statement 2 is the correct explanation of statement 1.

(B) Both statements 1 and 2 are correct but statement 2 is not the correct explanation of statement 1.

(C) Statement 1 is correct and statement 2 is incorrect.

(D) Both statements 1 and 2 are incorrect.

24. Which Column-I with Column-II and select the correct answer from codes given below.

Column I	Column II
A. ampr	(i) Artificial plasmid
B. macromolecular separation	(ii) Selectable marker
C. *Hind* III	(iii) Electrophoresis
D. pBR322	(iv) *Haemophilus influenza*

(A) A-(iii), B-(ii), C-(i), D-(iv)

(B) A-(iv), B-(i), C-(iii), D-(ii)

(C) A-(ii), B-(iii), C-(iv), D-(i) (D) A-(ii), B-(iv), C-(i), D-(iii)

25. Which of the following bacteria is used as a vector for plant genetic engineering?
(A) *Agrobacterium tumefaciens*
(B) *Bacteriophages*
(C) *Thermus aquaticus*
(D) *Pyrococcus furiosus*

26. Read the given statements and select the correct option.
Statement 1: The tumour inducing plasmid (Ti plasmid) acts as a cloning vector in recombinant DNA technology.
Statement 2: The Ti plasmid which is used in the mechanisms of delivering genes to a cell remains pathogenic.
(A) Both statements 1 and 2 are correct and statement 2 is the correct explanation of statement 1.
(B) Both statements 1 and 2 are correct but statement 2 is not the correct explanation of statement 1.
(C) Statement 1 is correct and statement 2 is incorrect.
(D) Both statements 1 and 2 are incorrect.

27.Which of the following is used to stimulate the formation of erythrocytes for patients suffering from anaemia?

(A) Calcitonin

(B) Erythropiotin

(C) Gonadotropin

(D) Interleukins

28. Match Column-I with Column-II with respect to the nomenclature of restriction enzyme *Eco* R I and select the correct answer from codes given below:

Column I	Column II
A. E	(i) Ist in order of identification
B. Co	(ii) Name of genus
C. R	(iii) Name of species
D. I	(iv) Name of strain

(A) A-(iii), B-(i), C-(ii), D-(iv) (B) A-(iv), B-(i), C-(iii), D-(ii) (C) A-(i), B-(ii), C-(iii), D-(iv)

(D) A-(ii), B-(iii), C-(iv), D-(i)

29. Read the given statements and select the correct option.

Statement 1: Restriction endonuclease enzymes recognize a specific palindromic nucleotide sequence in the DNA.

Statement 2: Restriction endonuclease enzymes are called as molecular scissors or biological scissors.

(A) Both statements 1 and 2 are correct and statement 2 is the correct explanation of statement 1.

(B) Both statements 1 and 2 are correct but statement 2 is not the correct explanation of statement 1.

(C) Statement 1 is correct and statement 2 is incorrect.

(D) Both statements 1 and 2 are incorrect.

30. Read the given statements and select the correct option.

Statement 1: The cloning vector is required to have very few, preferably single, recognition sites for the commonly used restriction enzymes.

Statement 2: Presence of more than one recognition sites within a cloning vector will generate several fragments, which will complicate the process of gene cloning.

(A) Both statements 1 and 2 are correct and statement 2 is the correct explanation of statement 1.

(B) Both statements 1 and 2 are correct but statement 2 is not the correct explanation of statement 1.

(C) Statement 1 is correct and statement 2 is incorrect.

(D) Both statements 1 and 2 are incorrect.

31. Read the given statements and select the correct option.

Statement 1: In recombinant DNA technology, human genes are often transferred into bacteria (prokaryotes) or yeast (eukaryotes).

Statement 2: Both bacteria and yeast multiply very fast to form huge populations which express the desired gene.

(A) Both statements 1 and 2 are correct and statement 2 is the correct explanation of statement 1.

(B) Both statements 1 and 2 are correct but statement 2 is not the correct explanation of statement 1.

(C) Statement 1 is correct and statement 2 is incorrect.

(D) Both statements 1 and 2 are incorrect.

32. The different steps of recombinant DNA technology are given below randomly.

(i) Isolation of DNA fragments or genes to be cloned.

(ii) Introduction of the recombinant DNA into a suitable cell (usually *E.coli*) called host (transformation)

(iii) Multiplication/expression of the introduced gene in the host.

(iv) Selection of the transformed host cells, and identification of the clone containing the desired gene/DNA fragment.

(v) Insertion of the isolated gene in a suitable plasmid vector.

Which of the following represents the correct sequence of steps?

(A) (i), (iii), (ii), (iv), (v) (B) (iii), (ii), (i), (v), (iv) (C) (i), (v), (ii), (iv), (iii) (D) (v), (i), (iii), (iv), (ii)

33. Fill up the blanks and select the correct option.

(i) *Eco* R I cuts the DNA between bases _____ only when the sequence _____ is present in the DNA.

(ii) Disruption of the cell membranes can be achieved by treating the bacterial cells, plant cells and fungal cells with enzymes respectively _____, _____ and _____.

(iii) Since, DNA has a _____ charge, it moves towards the _____ of the electrophoretic chamber.

(A) (i) G↓A, GAATTC (ii) endonuclease, cellulase, chitinase (iii) negative, anode

(B) (i) G↓A, GAATTC (ii) lysozyme, cellulase, chitinase (iii) positive, cathode

(C) (i) G↓A, GAATTC (ii) lysozyme, cellulase, chitinase (iii) negative, anode

(D) (i) G↓A, GAAATC (ii) lysozyme, cellulase, chitinase (iii) positive, cathode

34. Match Column-I (enzyme) with Column-II (Characteristic/activity) and select the correct answer from codes given below.

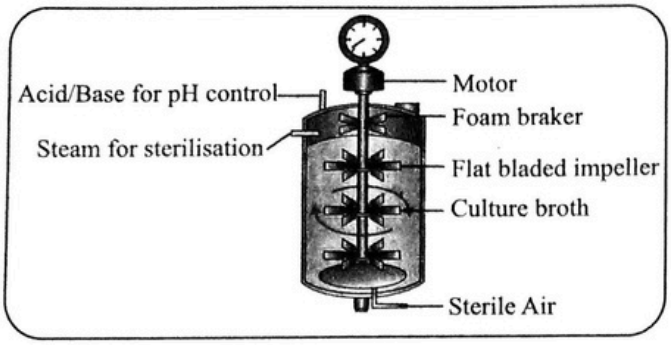
Column I	Column II
A. *Taq* DNA polymerase	(i) Cleaves the ends of linear DNA
B. Exonuclease	(ii) Breakdown of fungal cell membrane
C. Protease	(iii) Stable above 90°C
D. Chitinase	(iv) Made only by eukaryotic cells
	(v) Degradation of proteins

(A) A-(iii), B-(iv), C-(i), D-(ii) (B) A-(iv), B-(iii), C-(i), D-(ii) (C) A-(ii), B-(i), C-(v), D-(iii) (D) A-(iii), B-(i), C-(v), D-(ii)

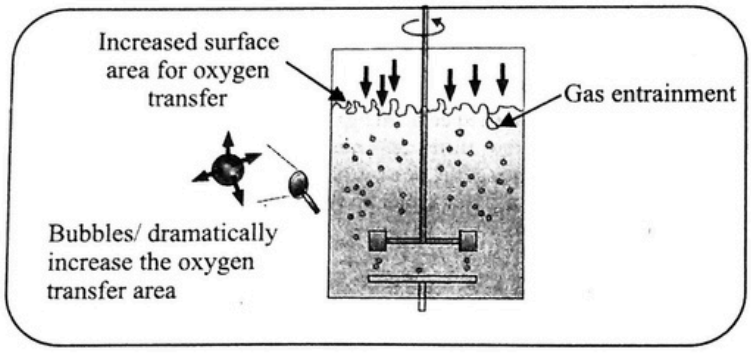
35. In which method of gene transfer the wall of the bacterium is made permeable by treating it with calcium chloride? [1]

(A) Electroporation
(B) Micro-injection
(C) Chemical- Mediated Genetic Transformation
(D) Gene gun

36. Identify the figures (A) and (B) and select the correct option.



(A)



(B)

	(A)	(B)
(A)	Sparged stirred-tank bioreactor	Simple stirred-tank bioreactor
(B)	Sparged stirred-tank bioreactor	Sparged stirred-tank bioreactor
(C)	Simple stirred-tank bioreactor	Sparged stirred-tank bioreactor

(D)	Simple stirred-tank bioreactor	Simple stirred-tank bioreactor

37. Enzyme '*Taq* polymerase' used in PCR, has been isolated from bacterium
    (A) *Agrobacterium tumefaciesn*
    (B) *Thermus aquaticus*
    (C) *Streptomyces albus*
    (D) *Escherichia coli*

38. In a polymerase chain reaction, temperature required for the steps
    (i) Denaturation, (ii) Annealing and
    (iii) Extension are respectively.
    (A) (i) 94°C (ii) 40°C (iii) 72°C
    (B) (i) 40°C (ii) 72°C (iii) 94°C
    (C) (i) 94°C (ii) 72°C (iii) 40°C
    (D) (i) 72°C (ii) 94°C (iii) 40°C

39. In addition to *Taq* polymerase enzyme which other thermostable DNA polymerases have isolated to be used in polymerase chain Reaction (PCR)?
    (A) *Pfu* polymerase isolated from *Pyrococcus furiosus*
    (B) *Tli* polymerase (vent polymerase) isolated from *Thermococcus litoralis*
    (C) Both A and B

(D) None of these

40. Which of the following statements are correct with respect to a bioreactor?

(i) It can process large volumes of culture.

(ii) It provides optimum temperature and pH.

(iii) It is completely an automated tool

(iv) It is a compact thermal cycler.

(A) (i) and (ii) (B) (i), (ii) and (iii)

(C) (iii) and (iv) (D) (ii) and (iii)

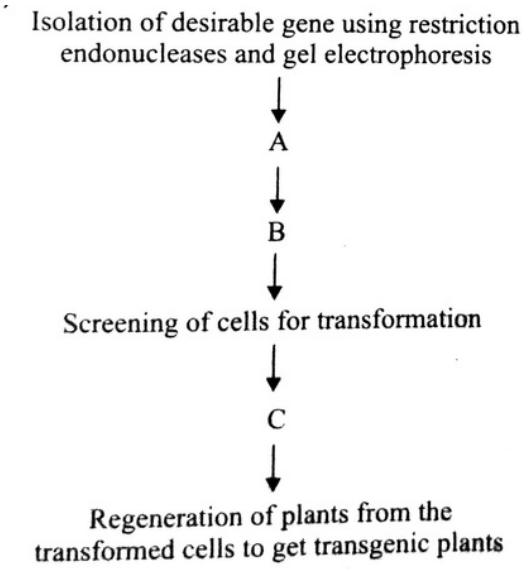
41. The given flow chart depicts the steps to transfer a desirable gene of interest into a plant.

Identify the missing steps (A, B and C) with regards to following statements and select the correct option.

(i) Joining of desirable gene to a suitable cloning vector using ligases to create a recombinant DNA molecule.

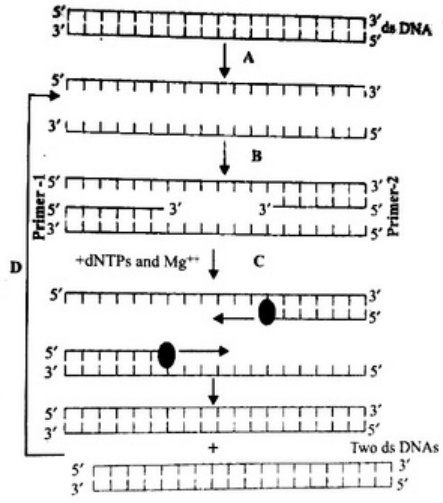
(ii) Selection of transformed cells.

(iii) Transferring the recombinant DNA molecules to the target cells.



A	B	C		A	B	C
(A) (i)	(ii)	(iii)		(B) (i)	(iii)	(ii)
(C) (ii)	(iii)	(i)		(D) (iii)	(i)	(ii)

42. Given figures represent the steps involved in polymerase chain reaction (PCR). Identify the steps A, B, C and D, and select the correct option.



	A	B	C	D
(A)	Denaturation at 94-96°C	Extension through *Taq* polymerase at 72°C	Repetition of denaturation and polymerization	Annealing at 72°C
(B)	Denaturation at 94-96°C	Annealing at 40 – 60°C	Extension through *Taq* polymerase at 72°C	Repetition of denaturation and polymerization
(C)	Denaturation at 40-60°C	Annealing at 72°C	Extension through *Taq* polymerase at 94-96°C	Repetion of denaturation and polymerization
(D)	Extension through *Taq* polymerase at 72°C	Denaturation at 94 – 96°C	Annealing at 72°C	Repetition of denaturation and polymerisation

43. Which one of the following is not a correct match?

(A) Tumour inducing - Ti plasmid

(B) DNA probing - Searching for desired DNA fragment

(C) PCR - DNA staining

(D) Agarose - Sea weeds

44. Study the following statements regarding recombinant DNA technology and select the incorrect ones.

(i) *Taq* polymerase extends the primers using the nucleotides provide in the reaction.

(ii) Antibiotic resistance genes are considered as desirable genes in recombinant DNA technology.

(iii) DNA fragments are separated according to their charge only, in agarose gel electrophoresis.

(iv) Transformation is a procedure through which a piece of DNA is introduced in a host bacterium.

(v) To produce higher yields of a desired protein, host cells can be multiplied in a continuous culture.

(vi) Downstream processing is one of the steps of polymerase chain reaction.

(A) (ii), (iii) and (vi)

(B) (i), (iii) and (v)

(C) (ii), (iii) and (v)

(D) (i), (v) and (vi)

45. Read the following statements and select the incorrect ones.

(i) The first transgenic buffalo, Rosie produced milk which was human alpha-lactalbumin enriched.

(ii) Restriction enzymes are used in isolation of DNA from other macromolecules.

(iii) Downstream processing is one of the steps of rDNA technology.

(iv) Disarmed pathogen vectors are also used in transfer of rDNA into the host.

(A) (ii) and (iii) (B) (iii) and (iv)

(B) (i) and (iii) (D) (i) and (ii)

ANSWERS

1. A 2. B 3. D 4. D 5. D 6. B 7. B 8. C 9. C 10.C
11. B 12. B 13. C 14. A 15. A 16. B 17. A 18. C 19.D
20.B 21. C 22. C 23. A 24. C 25. A 26. C 27. B 28. D
29.B 30.A 31. A 32. C 33. C 34. D 35. A 36. C 37. B
38.A 39.C 40.A 41. B 42. B 43. C 44. A 45. D

SOLUTIONS

1. Log phase shows rapid increase in number of cells whereas lag phase shows no significant increase in number of cells.
3. Biotechnology deals with techniques of using live micro-organisms, plant or animal cells or their components or enzymes from organism to produce products and processes (services) useful to human beings. In vitro fertilization, synthesis of recombinant gene, correcting a defective qene and developing a DNA vaccine are the parts of biotechnology.
6. HindII was the first discovered restriction endonuclease. It was isolated from Haemophilus influezae Rd. It produces blunt ends.

8. Recon is the smallest unit of DNA capable of recombination.

11. A and B are restriction endonucleases because same restriction enzyme cuts both foreign DNA and vector DNA at specific point. C is DNA ligase which joins

foreign DNA to vector DNA. The newly formed recombinant DNA is transformed in bacteria and the bacterial cells are allowed to divide.

12. DNA ligases are also called genetic gum. They join two individual fragments of double stranded DNA by forming phosphodiester bonds between them thus help in sealing of DNA fragments. Therefore acting as molecular glue. The enzyme used most often is T4 DNA ligase.

16. DNA is a negatively charged molecule.

27. Erythropiotin stimulates the formation of erythrocytes for patients suffering from anaemia during dialysis or in AIDS patients.

35. Electroporation is a method of gene transfer in which the wall of the bacterium is made permeable by treatment with calcium chloride or lysozyme.

37. The final step of PCR is extension, wherein Taq DNA polymerase (isolated form a thermophilic bacterium Thermus aquaticus) synthesizes the DNA region between the primers, using DNTPs (deoxyncleoside triphosphates) and Mg2+ the primers are extended towards each other so that the DNA segment lying between the two primers is copied. The optimum temperature for this polymerization step is 720 C. Taq polymerase remains a remains active during high temperature induced denaturation of double stranded DNA,

39. In addition to Taq DNA polymerase, Pfu polymerase and Tli polymerase have been isolated which are also

thermostable. Pfu polymerase is isolated from pyrococcusfuriosus. Til (vent) polymerase is isolated from Thermococcus litoralis.

44. Antibiotic resistance genes are selectable markers. Desirable genes are the ones which are introduced in the vector for getting desired protein product. In agarose gel electrophoresis, DNA fragments are separated according to their charge and size. For downstream processing.
45. In 1997, Rosie, the first transgenic cow was engineered to produce milk enriched with a human protein called alpha-lactalbumin, making it nutritionally more balanced. Restriction enzymes are used to cut DNA at specific sites.