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Class: 12
Subject: Biology
Topic: Biotechnology principles and processes
No. of Questions: 25

Q1. What would be the molar concentration of human DNA?

Sol. Average molecular wt. of nucleotide in human DNA is 130.86. The molecular wt. of human DNA will be. Therefore, 6×10^9 nucleotides (based on human genome project) × 130.86 = 784.56×10^9 gm/mol. The molar concentration of DNA can be calculated accordingly.

Q2. Besides better aeration and mixing properties, what other advantages do stirred tank bioreactors have over shake flasks?

Sol. Shake flasks are used for growing and mixing the desired materials on a small scale in the laboratory. A large scale production of desired biotechnological product is done by using 'bioreactors'. Besides better aeration and mixing properties, the bioreactors have following advantages:

a. Small volumes of cultures are periodically withdraw advantages:
b. It has a foam control system.
c. It has temperature and pH control systems.

Q3. Collect 5 examples of palindromic DNA sequences by consulting your teacher. Better try to create a palindromic sequence by following base pair rules.

Sol. Palindromic nucleotide sequences in the DNA molecule are groups of bases that form the same sequence when read both forward and backward. Five examples of palindromic DNA sequences are as follows:

a. 5' ------ G G A T C C --------- 3'
   3' ------ C C T A G G --------- 5'
b. 5' ------ A A G C T T ----------5'

   3' ------ T T C G A A ----------5'
c. 5' ------ A C G C G T --------- 3'
   3' ------ T G C G C A --------- 5'
d. 5'------ A C T A G T --------- 3'
e. 5' ------ A G G C C T --------- 3'
   3' ------ T C C G G A --------- 5'

Q4. Can you think and answer how a reporter enzyme can be used to monitor transformation of host cells by foreign DNA in additional to a suitable marker?

Sol. A reporter enzyme can be used to differentiate transformed cells by tracking down the activity of its cells so that they appear white in colour. The others, which appear blue in colour, indicate that cells do not carry foreign DNA.

Q5. Discuss with your teacher and find out how to distinguish between

a. Plasmid DNA and chromosomal DNA
b. RNA and DNA

Sol.

a. Plasmid DNA is a piece of symbiotic DNA present in addition to chromosomal DNA mostly in bacteria and yeast. It does not form the part of normal chromosomal DNA of the cell. Pleasmid DNA is capable of replicating independently.

b.

DNA	RNA
1. It is mainly confined to the nucleus. A small quantity occurs in mitochondria and chloroplasts. 2. Its quantity is constant in each cell of a species. 3. It contains deoxyribose sugar. 4. Its pyrimidines are adenine and thymine. 5. The amount of adenine is equal to the amount of thymine, also the amount of cytosine is equal to the amount of guanine. 6. It consists of 2 polynucleotide chains held together by hydrogen bonds, and coiled into a double helix. Some viruses ($\phi \times 174$) have single-stranded DNA. 7. Its molecular weight varies from to 6 million. 8. It is of 2 types : linear intranuclear and circular extranuclear. 9. It can replicate itself. 10. It controls structure, metabolism, heredity, differentiation and evolution. 11. It is a component of chromosomes.	1. It mainly occurs in the cytoplasm. A small quantity is found in the nucleus. 2. Its quantity varies in different cells. 3. It contains ribose sugar. 4. Its pyrimidines are adenine and uracil. 5. Adenine and uracil are not necessarily in equal amounts, nor are cytosine and guanine 6. It consists of a single polynucleotide chain. It may fold on itself and get hydrogen-bonded and coiled into a pseudohelix. Some viruses (rheovirus) have double-stranded RNA. 7. Its molecular weight varies from 25,000 to 2 million. 8. It is of 3 types: mRNA, tRNA, rRNA. Each type is further of may subtypes. 9. It cannot replicate itself. It is formed by DNA. Some RNA viruses (paramyxo virus) can produce RNA from RNA template. 10. It brings about protein synthesis. It also starts replication.

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12. It is a genetic material in all organisms. 13. It dones not contain unusual bases. 14. A primer is needed for replication. 15. Its renaturation after melting is slow. 16. Its transfers its information to mRNA (transcription). 17. DNA is hydrolysed by the enzyme DNA-ase.	11. It is a component of ribosomes. 12. It is a genetic material in certain viruses. 13. It may contain unusual bases in addition to the normal ones. 14. No primer is needed for transcription. 15. Its renaturation after melting is quick. 16. mRNA transfers its information to polypeptide (translation). 17. RNA is hydrolysed by the enzyme RNA-ase.

Q6. Define biotechnology.

Sol. Biotechnology is the scientific manipulation of biological, especially at the molecular level, to produce new products for beneficial use.

Q7. Name a 'natural genetic engineer' of plants.

Sol. Agrobacterium tumefaciens, a crown gall bacterium is called natural genetic engineer of plants.

Q8. Name the enzyme involved in the continuous replication of DNA strand. Mention the polarity of the template strand.

Sol. Restriction enzyme: DNA dependent DNA polymerase catalyses polymerization only in one direction, i.e., 5' ⟶ 3'. This creates some additional complications at the replication fork. On one strand, replication is continuous and on the other discontinuous.

Q9. What are palindromic nucleotide sequences?

5'-------- G A A T T C ------- 3'
3' ------- C T T A A G --------5'

Sol. These are groups of nucleotides that form the same

Words when read both forward and backward. For example –

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Q10. Explain the contribution of Thermus aquaticus in the amplification of a gene of interest.

Sol. Amplification of gene is carried out by the technique called Polymerase Chain Reaction to synthesise multiple copies of the desired gene. This technique is based on annealing at 40–60^{o}C and synthesis of new strand, i.e., extension and denaturation at high temperature, i.e., 70–90^{o}C. High temperature may inactivate the DNA polymerase. Therefore, a thermostable (i.e., heat stable) Taq DAN polymerase extracted from heat stable bacterium Thermus aquaticus is used to carry out the process of amplification without any difficulty and without adding new enzyme to the PCR process.

Q11. What are recombinant proteins? How do bioreactors help in their production?

Sol. A piece of foreign DNA, once established inside the bacterial, plant or animal cell by the genetic engineering technique, starts expressing its role in the synthesis of protein. Such a protein, synthesized by recombinant DNA in a heterologous host, is called recombinant protein. These cells having rDNA are grown on a small scale in the laboratory to obtain recombinant protein. However, large scale production of the product is carried out in bioreactors.

Q12. How is DNA isolated in purified form from a bacterial cell?

Sol. The bacterial cells are treated with lysozyme to break the cells open and release macromolecules. Purified form of DNA is obtained by removing RNA by treatment with ribonuclease, proteins by treatment with protease and other molecules by appropriate treatments. Purified DNA is precipitated out by addition of child ethanol.

Q13. Name two commonly used bioreactors. State the importance of using a bioreactor.

Sol. The two most commonly used bioreactors are simple stirred-tank bioreactor and sparged stirred-tank bioreactor. The importance of using bioreactors is as follows:

a. It provides large volume for cultures. Thus, products are obtained in high quantity.
b. It provides. Optimal temperatures and pH for growth of desired product.

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Q14. A. Explain how to find whether and E. coli bacterium has transformed or not when a recombinant DNA brearing ampicillin resistant gene is transferred into it.

B. What does the ampicillin resistant gene act as in the above case?

Sol.

a. The successful transformation of E. coli with a recombinant DNA can be detected by growing the E. coli cell on culture media having amplicillin. The transformed cells will survive as they carry recombinant DNA with ampicillin resistance gene. Where as non-transformed cells will die s they are ampicillin sensitive.
b. Antibiotic resistance genes such as ampicillin resistance, serve as electable markers as in the above case.

Q15. Why and how bacteria can be made 'competent'?

Sol. The bacterial cells are made competent by retain them with a specifc concentration of divalent cations like calcium or magnesium e.g., C_aCl_2. The cells are then incubated with recombinant DNA on ice, followed by heat shock and then again ice. This makes the cell wall permeable and bacterial cell take up the plasmid DNA.

Q16. Name the source of the DNA polymerase used in PCR technique. Mention why it is used.

Sol. The most commonly used matrix is agarose which is a natural polymer extracted from sea weeds. The DNA fragments separate according their size through sieving effect provided by the agarose gel.

Q17. Write any four ways used to introduce a desired DNA segment into a bacterial cell in recombinant technology experiments.

Sol. Recombinant DNA technology involves the following steps:

a. Isolation of desired DNA fragment.
b. Fragmentation of DNA at specific locations by enzymes.
c. Amplification of recombinant DNA using PCR.
d. Insertion of recombinant DNA into the host cell.

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Q18. What is the principle of PCR?

Sol. The basic principle of PCR is that the double stranded DNA molecule, when heated to a high temperature, the two strands separate yielding single-stranded DNA molecules. The single-stranded DNA molecules easily be copied with the help of a DNA polymerase and nucleosides resulting in the duplication of original DNA molecule. By repeating these events, multiple copies of the original DNA molecule can be generated.

Q19. Write the major steps involved in gene cloning.

Sol. The technique of gene cloning involves – (i) Isolation of desired DNA fragment containing the gene to be cloned: (ii) Isolation of the vector used as cloning vehicle: (iii) Incubation of DNA fragments and digested vector in presence of DNA ligase producing recombinant DNA molecules: and (iv) Introduction of recombinant DNA into the host.

Q20. Do you see the prospects of viroids being used as plant vectors in near future?

Sol. Viroids are smallest and simplest pathogenic agents comprising of 300-400 bases long, circular, single stranded, naked RNA. They are mechanically transferable and infect other parts of the plants. They have capability of being used as plant vectors.

Q21. Which part would be most suitable for raising virus-free plants for micropropagation?

a) Meristem

b) Node

c) Bark

d) Vascular tissue

Sol. a)

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Q22. For transformation, micro-particles coated with DNA to be bombarded with gene gun are made up of

a) Silicon or Platinum

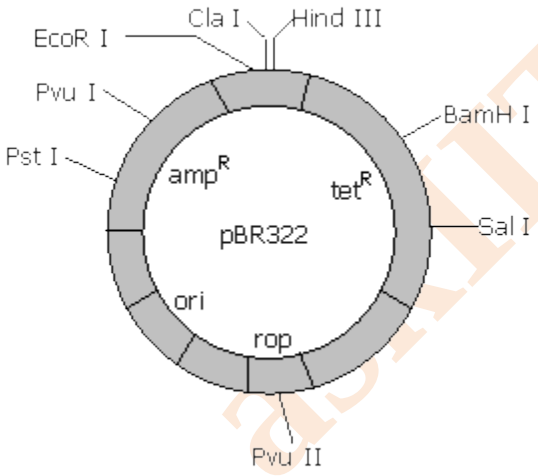
b) Gold or Tungsten

c) Silver or Platinum

d) Platinum or Zinc

Sol. c)

Q23. The figure below is the diagrammatic representation of the E.coli vector pBR 322. Which one of the given options correctly identifies its certain component(s)?



a) Hind III, EcoRI-selectable markers

b) ampR, tetR-antibiotic resistance genes

c) ori-original restriction enzyme

d) rop-reduced osmotic pressure

Sol. (b)

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Q24. In plant biotechnology, PEG is used in

a) Protoplast fusion

b) Cell culture preparation

c) Protoplast isolation

d) Hardening

Sol. (a)

Q25. Polyethylene glycol method is used for

a) Energy production from sewage

b) Gene transfer without a vector

c) Biodiesel production

d) Seedless fruit production

Sol. (b)