Class: XII
Biotechnology (045)
Marking Scheme 2018-19

Time allowed: 3 Hours **Maximum Marks: 70**

	SECTION A	
1.	$4x2^{20}$ OR The LEU2 gene codes for an enzyme which is needed for the synthesis of the amino acid leucine. Yeast cells having this plasmid can grow on a medium lacking leucine and hence can be selected over cells not containing the plasmid.	**1**
2.	Removal of terminal phosphate group at the 5' end of DNA prevents self-ligation of vector DNA.	**1**
3.	As specific activity increases, purity also increases.	**1**
4.	No, animal cells need a CO_2 incubator.	**1**
5.	Selectively removing a gene and making other precise genetic modifications in the mouse ES cells and creating mouse models of human disease.	**1**
6.	Mad cow disease caused by prion/ rogue protein. OR Protein Efficiency Ratio PER is used as a measure of growth expressed in terms of weight gain of an adult by consuming 1g of food protein.	**1**
	SECTION B	
7.	Inverted microscopes have objectives at bottom unlike regular microscopes. (Light source is at top) Animal cells are present at the bottom of culture vessels and hence easier to observe. OR Advantages of animal cell culture: Homogenous genetic population, Controlled physico-chemical environment, Easy to add genes, Available in adequate numbers, Easy production, No ethical clearance required, Cost effective. (any two of the above points) Disadvantages of animal cell culture: small size sensitive to detect the changes, challenging scale up, may not represent in vivo phenotype and genotype. (any two of the above points)	**2**
8.	Insertional inactivation of the lac Z gene present on the vector (e.g. pUC 19). This gene expresses the enzyme β-galactosidase whose activity can cleave a colourless substrate called X-Gal into a blue coloured product. If the lac Z gene is inactivated due to the presence of the insert then the enzyme is not expressed. Hence after the transformation experiment recombinants will appear as white colonies and non-recombinants appear as blue colonies.	**2**
9.	Interspecific crosses lead to abnormal endosperm development resulting in premature death. Embryo should be excised and cultured	**2**

10.	Two dimensional gel electrophoresis. The proteins are separated by their isoelectric points (pI) in the first dimension (IEF), and according to their molecular mass in the second dimension (SDS-PAGE). Cumulative effects of these techniques lead to high resolution	**2**
11.	It shows inaccuracy in gene prediction. Secondly there is no correlation between the intuitive complexity of an organism Yeast encodes 70 percent of proteins whereas worm and fruit fly encode 20-25% OR Fig 7, pg 72; A textbook of Biotechnology.	**2**
12.	Culture volume= 800 l Number of animal cells= 10^5 x 1000x 800= 8x 10^{10} Number of virus particles=50x8x10^{10} =4x10^{12} 10^6 (mol.wt) has 6.023 x10^{23} virus particles. Hence 4x10^{12} particles have weight 4x10^{12}x10^6/6.023 x10^{23} = 0.667x 10^{-5} gm Volume of one virus particle= 4/3π r^3= 4/3x22/7x1^3 nm^3 Volume of 4x10^{12} virus particles =1.67x 10^{13} nm^3 OR (a) Insulin production is 100 mg/L; so fermentor volume needed for 1 Kg of insulin is 1 Kg / 100mg = 1000, 000mg/100,g = 10,000mg = 10,000L. So we need 10,000-litre fermentor to produce 1 Kilogram of insulin in one batch. (b) In this case the cell concentration is increased to 50 g/L; so insulin production per liter will be 50 X 100 = 5000 mg = 5 g / L; Thus, to produce 1 Kilogram of insulin we need 1 Kilogram / 5 g = 1000 g / 5g = 200 g. So, if the cell concentration is increased 50 times, we need 200-litre reactor to produce 1 Kilogram of insulin.	**2**
13.	Somaclonal variations (a)It is associated with chromosomal variation (b) It helps in production of mutants e.g. disease resistant in Potato (any 1)	**2**
14.	Causes: 1.presence of proteins in the culture medium 2. some molecules produced by the microbes. Control: Antifoams such as fatty acids(olive oil, sunflower oil) or silicone (any 1)	**2**
	SECTION C	
15.	Transfer of genes into ova/stem cells of animals with a view to obtain large scale production of the proteins encoded by these genes in the milk, urine or blood of such animals. Advantages- 1.High Production capacity 2. Ease of collection of source material 3/ Moderate capital instrument requirements and low operational cost 4. Ease of production, scale up	**3**

16.	Figure 11, Page 151; A Text book of Bio-Technology Class XII	**3**
17.	(a)Southern hybridization (b) i. Digestion of genomic DNA with restriction enzymes ii. Electrophoresis on agarose gel iii. Transfer to a nitrocellulose membrane (Southern Blotting) iv. Hybridization with labeled probe v. Detection of labeled probe. Or through fig 10, pg 20; A textbook of Biotechnology CBSE Class XII	**3**
18.	It is due to SNP that is single nucleotide polymorphism It is estimated that differences between human and chimpanzee is only 1-3% while human and mouse share about 97.5% of their working DNA OR Macromolecular Sequences first compiled in this Atlas Development of computer methods for comparison of protein sequences. Detection of various features from sequences like duplications, evolutionary histories, alignments etc.	**3**
19.	Diagram of mass spectrometer as on pg 45, Fig 10; A Textbook of Biotechnology Protein sequences/ Molecular Mass can be determined. OR (i) kappa casein contains a lipid molecule. 2/3 of the protein is hydrophobic (ii) Whey protein results in elevation of a tripeptide glutathione in cells which detoxifies xenobiotics. (iii) Curd is a good source of beneficial bacteria which can colonise the intestinal tract.	**3**
20.	Naturally derived or synthetic materials may be engineered into "scaffolds" that when implanted in the body could provide a template that allows the body's own cells to grow and form new tissues. Such implants could function like neo-organs in patients without triggering immune responses. Genetically-modified animals may also provide a source of cells, tissues, and organs for xenografts Applications 1.The aim of tissue engineering is to supply body parts for repair of damaged tissue and organs, without causing an immune response or infection or mutilating other parts of the body. 2.Tissue engineering potentially offers dramatic improvements in low-cost medical care for hundreds of thousands of patients annually. 3. Largescale culturing of human or animal cells-including skin, muscle, cartilage, bone, marrow, endothelial and stem cells-may provide substitutes to replace damaged components in humans. (Any two) OR A growth curve can be established from which one can obtain a population doubling time, a lag time, and a saturation density. A growth curve generally will show the cell population's lag phase, that is, the time it takes for the cells to recover from subculture, attach, and spread; the log phase, in which the cell number begins to increase exponentially and a plateau phase, in which the growth rate slows or stops due to depletion of growth factors and nutrients. An increase in cell number is also a frequently used method of assessing the effect of hormones, nutrients, and so forth on	**3**

	a specific cell type Fig 4 , pg 144 ; A textbook of biotechnology	
21.	i. Persons must wash their hands with germicidal soap after handling viable microorganisms. ii. Eating, drinking, smoking etc. are not permitted in the working area. iii. Food is to be stored outside the work area in cabinets / refrigerators designated for this purpose only. iv. Mouth pipetting is strictly prohibited. v. All procedures are carried out in a way, so as to minimize splashes and generation of aerosols. vi. After work with viable microorganisms, work surfaces are decontaminated thoroughly. vii. It is recommended that laboratory coats / aprons should be worn while working. viii. All cultures, stocks or other waste are decontaminated and autoclaved before disposal.(Any six)	**3**
22.	(a) Blunt ends : *Alu I* 5'-A-G-C-T-3' → 5'-A-G + C-T-3' 3'-T-C-G-A-5' → 3'-T-C + G-A-5' (b) Sticky ends: *Eco RI* 5'-G-A-A-T-T-C-3' → 5'-G-3' + 5'-A-A-T-T-C-3' 3'-C-T-T-A-A-G-5' → 3'-C-T-T-A-A-5' + 3'-G-5' Cohesive or sticky ends are self-annealing and easier to ligate in making recombinant vectors.	**3**
23.	1. The safety of GM food for human and animal consumption (e.g. GM food may cause allergenicity). 2. The effect of GM crops on biodiversity and environment. The effect of GM crops on non-target and beneficial insects/microbes. 3. Transgenes may escape through pollen to related plant species (gene pollution) and may lead to the development of super weeds. 4. The GM crops may change the fundamental vegetable nature of plants as the genes from animals (e.g. fish or mouse) are being introduced into crop plants. 5. The antibiotic resistance marker genes used to produce transgenic crops may horizontally transfer into microbes and thus exacerbate problem of antibiotic resistance in human and animal pathogens (i.e. transgenes may move from plants to gut microflora of humans and animals). 6. The GM crops may lead to the change in the evolutionary pattern.(Any three)	**3**
24.	Metagenomics approach has been developed to identify and select microbial genes synthesizing novel molecules. This approach directly utilizes the large number of microbial genomes present in an environmental niche, for example in soil, in water such as ocean or in human gut. These genomes are contributed by both the culturable and the nonculturable variety of microbes and together constitute what has been termed as metagenome.	**3**

	The collective DNA is extracted from a sample of soil, water or any other environmental niche. It is subjected to restriction digestion using restriction endonucleases and the fragments are cloned into suitable vectors. The clones are then screened for presence of a variety of molecules. The clones expressing novel molecules or molecules with improved characteristics are used for large-scale production by fermentation techniques. Metagenomics approach has been developed to identify and select microbial genes synthesizing novel molecules. Fig 11, pg 103; A Textbook of Biotechnology Class XII	
25.	Plants raised by tissue culture of somatic hybrid cells, formed by fusion of plant cell protoplasts are called as somatic hybrids. Procedure: Isolation of plant cell protoplasts, their fusion, selection of hybrid cells and raising by plant tissue culture OR Herbicide and other chemicals affect the crops which can be resolved by introducing modified gene for the over production of herbicide target enzyme This enzyme expresses in chloroplast in plants So that it becomes insensitive to the herbicide	**3**
	SECTION D	
26.	3'OH group is absent in ddNTP's which cause termination of growing DNA chain during Sanger's DNA sequencing method. DNA fragments formed by chain termination in all the four tubes for the given strand 3' ATGCTAGC 5' OR (i)BAC and YAC- BAC- Insert size-50-500 kb YAC-Insert size-250-1000 kb (ii) pBR322 and pUC 19-pBR322- contains two antibiotic resistance genes , pUC 19- contains Lac Z gene (iii) M-13 and lambda phage- M-13- circular single stranded, lambda phage- linear double stranded (iv) Cosmid and Plasmid Cosmid- having features of plasmid and cos sites of phage lambda, insert size 30-40Kb Plasmid- small, circular, extra-chromosomal, self -replicating, naturally present in bacteria insert size 0.5-8 Kb **(v)**Transformation and Transfection- Transformation- Cold $CaCl_2$ treated competent bacterial cells to introduce Rdna Transfection-Transfer rDNA in to host cells by mixing foreign DNA with charged substances like calcium liposomes/ calcium phosphate/ DEAE dextran	**5**
27.	Primary, secondary, tertiary and quarternary structure with details (pg 31; A Textbook of Biotechnology Class XII) Covalent- disulfide bonds Non covalent- Hydrogen, Van der waals forces of attraction, Hydrophobic and Ionic	**5**

bonds
Pg 32; A Textbook of Biotechnology Class XII

OR

Aqueous two phase partition with diagram (fig 8) on pg 42; A Textbook of Biotechnology Class XII

Stabilising steps

1. Maintenance of a specific pH value range of buffered solutions in which a protein is maximally stable.
2. Maintenance of physiological conditions (%CO_2 for animal cell culture and temperature).
3. Use of inhibitors to prevent the action of proteolytic enzymes.
4. Avoidance of agitation or addition of chemicals which may denature the target protein.
5. Minimise processing time. (Any three)

28. **5**

Single-gene mutations which follow mendelian inheritance	**Gene polymorphisms which has complex inheritance**
Cystic Fibrosis (Cystic Fibrosis Transmembrane Conductance Regulator CFTR gene) 1. Inheritance: autosomal recessive disease 2. Genomic location: Chromosome 7 (7q31.2) 3. Mutation: The most common mutation is a deletion of 3 bps resulting in the loss of codon no. 508, which codes for phenylalanine	**Common late-onset Alzheimer's disease** 1. Inheritance: Major cause is epsilon4 allele of the gene coding for apolipoproteinE (APOE) 2. Genomic location: Chromosome 19 (19q13) and recently Chromosome 10 (10q21).
Huntington disease (Huntingtin gene HTT) 1. Inheritance: autosomal dominant 2. Location: Chrosome 4 (4p16.3) 3. Mutation: increased number of CAG repeats more than 35 times	**Migraine** 1. Susceptibility locus: Chromosome 6p12.2 - 6p21.1 and Chromosome 1q31

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