

**ANIMAL CELL CULTURE & ANIMAL BIOTECHNOLOGY  
(BIOT 4101)**

Time Allotted : 3 hrs

Full Marks : 70

*Figures out of the right margin indicate full marks.*

*Candidates are required to answer Group A and any 5 (five) from Group B to E, taking at least one from each group.*

*Candidates are required to give answer in their own words as far as practicable.*

**Group – A  
(Multiple Choice Type Questions)**

1. Choose the correct alternative for the following: **10 × 1 = 10**

- (i) For culturing, plasma from the adult chicken is preferred to mammalian plasma because
- (a) it forms a clear and solid coagulum even after dilution
  - (b) it is too opaque
  - (c) it doesn't produce solid clots
  - (d) it forms a semi solid coagulum.

- (ii) Match between the cell properties (group-I) and cell assay (group-II)

Cell properties (Group-I)	Assay (Group-II)
(P) Enzymatic property	(1) Dye exclusion
(Q) Membrane integrity	(2) Clonogenic assay
(R) Cell division	(3) MTT assay
(S) Colony forming potential	(4) Cell counting

Which one of the following is the correct match between group-I and group-II?

- (a) P-2; Q-4; R-1; S-3
  - (b) P-3; Q-1; R-4; S-2
  - (c) P-1; Q-3; R-4; S-2
  - (d) P-3; Q-2; R-4; S-1.
- (iii) Disaggregating of cells can be achieved by
- (a) physical disruption
  - (b) enzymatic digestion
  - (c) treating with chelating agents
  - (d) all of the above.
- (iv) In protein production using mammalian cells, polyethyleneimine (PEI) (Cation polymers) are used for:
- (a) Transfection with plasmid
  - (b) Supplementing the SD-PAGE running buffer
  - (c) Protein determination
  - (d) RNA extraction.

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- (v) Excess CO<sub>2</sub> suppress cell growth and productivity by  
(a) inhibiting respiration  
(b) altering intracellular pH by diffusing across cell membrane  
(c) both (a) and (b)  
(d) altering pH of the medium.
- (vi) Identify the cell culture medium below that normally contains phenol red as an additive to detect pH changes.  
(a) LB medium  
(b) Cell culture medium for sf9 cells  
(c) YPD yeast cell culture medium  
(d) Cell culture medium used for mammalian cells.
- (vii) The cell line used for the production of recombinant polio vaccine is  
(a) Dog kidney cell line  
(b) CHO cell line  
(c) Primate kidney cell line  
(d) Mouse fibroblast cell line.
- (viii) Which of the following statements is always correct about bioreactor?  
P. Substrate concentration inside the bioreactor is equal to that in the exit stream.  
Q. Optima dilution rate is lower than critical rate.  
R. Biomass concentration increase with increase in dilution rate.  
S. Cell recirculation facilitates operation beyond critical dilution rate.  
(a) P and Q only  
(b) P, R, and S only  
(c) P and S only  
(d) P, Q and S only.
- (ix) Yield coefficient  $Y_{x/s}$  is defined as  
(a)  $(x - x_0)/(s_0 - s)$   
(b)  $(s_0 - s)/(x - x_0)$   
(c)  $(x_0 - x)/(s - s_0)$   
(d)  $(s - s_0)/(x_0 - s)$ .
- (x) A T-flask is seeded with  $10^5$  anchorage-dependent cells. The available area of the T-flask is 25 cm<sup>2</sup> and the volume of the medium is 25 ml. Assume that the cells are rectangles of size 5 $\mu$ m  $\times$  2 $\mu$ m. If the cells grow to monolayer confluence after 50 h, the growth rate in number of cells/(cm<sup>2</sup>.h) is  
(a)  $2.5 \times 10^5$   
(b)  $25 \times 10^5$   
(c)  $2.0 \times 10^5$   
(d)  $5.2 \times 10^2$ .

### Group – B

2. (a) Write the names of different parameters required for optimal growth of animal cell culture.
- (b) What are basal media and complete media? Give one example of each.
- (c) Write the names of different serum components and their role in animal cell culture. Why heat inactivation of serum is done before using in the media? Why serum can't be frozen after heat inactivation?

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- (d) Write the names of three animal cell lines. Write three suitable suppliers name for getting animal cell lines.

$$2+(2 + 1) + (3 + 1 + 1) + (1 + 1) = 12$$

3. (a) How animal cells are stored in the laboratory?  
(b) Discuss the method for thawing and recovering of animal cells from a frozen sample.  
(c) How you can determine the viability and cell number using hemocytometer and trypan blue staining?

$$4 + 4 + 4 = 12$$

### Group – C

4. (a) Define dilution factor.  
(b) Mention the basic components of animal cell culture bioreactor.  
(c) Explain the basic parameters to be controlled inside an animal cell culture bioreactor.

$$2 + 3 + 7 = 12$$

5. (a) Why CO<sub>2</sub> is needed for animal cell culture?  
(b) Animal cells are growing having doubling time  $t_d = 2.5$  hrs. The saturation constant for this substrate  $K_s = 1.2$  kg/m<sup>3</sup> and the yield coefficient  $Y_{X/S} = 0.42$ . For a chemostat operating with a feed conc. of 35 kg/m<sup>3</sup> ( $S_0$ ) of blood serum, calculate the following:  
(i) A cell concentration, when the dilution rate  $D$  is one half of  $D_{max}$  output.  
(ii) Substrate conc., when the dilution rate  $D$  is 0.75,  $D_{max}$ .  
(iii) Maximum dilution rate for maximum output of cells.  
(iv) Cell productivity,  $X D$  at  $D = 0.75 P_{max}$  output.

$$2 + 10 = 12$$

### Group – D

6. (a) Write the names of two techniques of animal cloning. Describe one technique for animal cloning which is based on stem cell technology.  
(b) Describe the technique of production of knock-out mice using discovery of homologous recombination, which brought Nobel Prize in Physiology and Medicine in 2007 to Mario R. Capecchi, Sir Martin J. Evans and Oliver Smithies.  
(c) "Transgenic animal are bioreactor for large scale production of substances of human welfare and improvement of production of traits". Justify the statement with examples.

$$(1 + 4) + 4 + 3 = 12$$

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7. (a) Describe the steps of IVF with labeled diagram.  
(b) What is chimera? Define gene targeting and gene trapping.  
(c) Describe four types of Inhibitory "Anti-Gene" expression strategies as therapeutic agents.

$$3 + (1 + 2 + 2) + 4 = 12$$

**Group – E**

8. (a) Using only a labelled diagram describe the steps of ES cell collection.  
(b) Describe the approaches of stem cell therapies for neurological disease using transplantation of neural stem cells.  
(c) Explain with examples the differences between biopharmaceuticals and biosimilars.  
(d) With labelled diagrams describe the steps of Baculovirus and insect cell expression system for biomanufacturing of biopharmaceuticals.

$$3 + 3 + 2 + 4 = 12$$

9. (a) Describe the generation of human induced pluripotent stem cells for use in cell therapy.  
(b) Describe the application of stem cells for the treatment of hemophilia and diabetes mellitus.  
(c) Describe with labelled diagram the process of cell fusion by virus mediated and electrofusion method in hybridoma technology.

$$3 + 3 + (3 + 3) = 12$$

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