7. (a) What is decimal reduction time for sterilisation of media? How is it related to specific death constant of organism?

(b) It is desired to pasteurise 240 litre/min milk in a continuous pasteuriser by heating to 71°C for a sufficient time to achieve 12 D sterilization. An existing pasteuriser comprising of well insulated pipe (55 mm ID and 30 m length) fed from a plate heat exchanger is available for the duty. Will it provide sufficient holding time? K_d for the organism is 1.84 sec^{-1}.

(2 + 4) + 6 = 12

Group - E

8 What fraction of the cells undergoing division will generate a plasmid free cell if

(i) all cells having 40 plasmid at division?

(ii) all cells have enough plasmid DNA for 40 copies, but one half of the plasmid DNA is in the form of dimer and $1/5^{th}$ in the form of tetramer?

(iii) half of the cells have 10 copies of the plasmid and half have 70 copies (the average copy number is 40 as in case (i)?

4 + 4 + 4 = 12

9. Describe the process of industrial production of lactic acid with a flow diagram.

12

BIOPROCESS TECHNOLOGY
(BIOT 5203)

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) Immobilization of enzymes helps to
   (a) recycle the enzyme
   (b) recover the enzyme easily
   (c) reduce downstream processing cost
   (d) all of the above.

(ii) Both K_m and V_{max} are affected by the inhibition factor in which type of enzyme inhibition?
   (a) Competitive   (b) Non-competitive
   (c) Uncompetitive   (d) All of the above.

(iii) Amount of CO_2 produced per unit amount of O_2 consumed is known as
   (a) yield coefficient   (c) respiratory quotient
   (d) degree of reduction   (d) maintenance coefficient

(iv) Which of the following method of sterilization has no effect on spores?
   (a) Drying   (b) Hot air oven
   (c) Autoclave   (d) None of these.

(v) The lowest temperature that kills all microorganisms in a liquid suspension in 10 minutes is known as the
   (a) decimal reduction time   (b) thermal death point
   (c) thermal death temperature   (d) thermal death time.

(vi) A batch reactor is a
   (a) steady state reactor   (b) unsteady state reactor
   (c) ideal reactor   (d) none of the above.

(vii) Balanced growth occurs in
(a) lag phase   (b) log phase
(c) stationary phase   (d) death phase.

(viii) Immobilization by surface immobilization results in
(a) washing out of enzymes
(b) minimum diffusion barrier between enzyme and substrate
(c) strong linkage between enzyme, substrate and matrix
(d) both (a) and (b).

(ix) Which of the following cannot be used as vector?
(a) Phase   (b) Plasmid
(c) Bacterium   (d) All can be used as vector.

(x) The enzyme used in the polymerase chain reaction is
(a) restriction endonuclease   (b) reverse transcriptase
(c) DNA polymerase   (d) RNA polymerase.

Group-B

2. Decarboxylation of glyoxalate(S) by mitochondria is inhibited by malonate(I). The following data were obtained in batch experiments:

S, mM		0.25	0.33	0.4	0.5	0.6	0.75	1.00
V, mM/h	I=0	1.02	1.39	1.67	1.89	2.08	2.44	2.5
V, mM/h	I=1.26mM	0.73	0.87	1.09	1.3	1.41	1.82	2.17
V, mM/h	I=1.95mM	0.56	0.75	0.85	1	1.28	1.39	1.82

(i) What type of inhibition is this?
(ii) Determine the constants V_{max}, K_m' and K_I.

$$2 + 10 = 12$$

3. (a) Derive a kinetic equation for non-competitive inhibition.

(b) The following data were obtained for an enzyme catalyzed reaction. Determine V_{max} and K_m' by inspection. Plot the data using Eadie-Hofstee method and determine these constants graphically. Explain the discrepancy in your two determinations. The initial rate data for the enzyme catalyzed reaction are as follows:
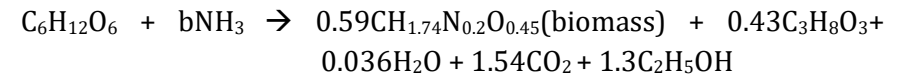
[S], μmol/L	500	200	60	40	30	20	16	10	8
v,μmol/L.min	125	125	121	111	96.5	62.5	42.7	13.9	7.5

$$4 + 8 = 12$$

Group-C

4. (a) Pseudomonas sp has a mass doubling time of 2.4h when grown in acetate medium. The saturation constant is 1.3g/l. The cell yield is 0.46g/g. If we operate a chemostat on a feed stream containing 38g/l acetate, find the following:
   i. maximum dilution rate
   ii. cell concentration when the dilution rate is one-half of the maximum.
   iii. substrate concentration when the dilution rate is $0.8D_{max}$.
   iv. cell productivity at $0.8D_{max}$.

(b) Derive an equation to determine the relationship of dilution rate to specific growth rate for a chemostat.

$$7 + 5 = 12$$

5. The growth of baker's yeast on glucose under anaerobic conditions can be described by the following overall reaction:

$$C_6H_{12}O_6 + bNH_3 \rightarrow 0.59CH_{1.74}N_{0.2}O_{0.45}(biomass) + 0.43C_3H_8O_3+$$
$$0.036H_2O + 1.54CO_2 + 1.3C_2H_5OH$$

   i. Determine $Y_{x/s}$.
   ii. Determine the yield coefficients: $Y_{EtOH/S}$, $Y_{CO2/S}$ and $Y_{C3H8O3/S}$.
   iii. Determine the degree of reductions for substrate, bacteria and RQ for the organism
   iv. Determine the coefficient 'b'.

$$1 + 3 + 6 + 2 = 12$$

Group - D

6. The time temperature data of heating and cooling periods of a 40,000 lit tank during sterilization of medium is :

Time (min)	0-15	15-40	40-50	50-60	>60
Temperature °C	85	121	85	55	30

The medium contains vitamins whose activation energy for destruction is 10kcal/gmol and the value of Arhenious constant is $10^4 min^{-1}$. Assume vitamin destruction is of first order and its initial concentration is 50 mg/lit. R is 1.99 cal/gmol°K. The medium contains 2.5×10^3 spores/lit. The spores have an activation energy 65 Kcal/gmol and K_d at 121°C is 1.02 min⁻¹.

(i) Estimate The probability of successful sterilization
(ii) What fraction of vitamin remains active?

$$6 + 6 = 12$$