M.TECH/BT/2ND SEM/BIOT 5203/2015
2015

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 x 1=10**

(i) 'Del Factor' is represented as

(a) $\ln N_o/N_t$ (b) A. $e^{-E/RT}$ (c) E/RT^2 (d) kN.

(ii) D value in sterilization of fermentation media is known as

(a) Time to kill 90% of organisms (b) Thermal death temperature
(c) Time to kill 10% of organisms (d) Thermal death point.

(iii) A higher *Km* value of Michaelis- Menten equation means

(a) greater affinity of enzyme to substrate
(b) lower affinity of enzyme to substrate
(c) unaffected with substrate binding
(d) lower dissociation constant value.

(iv) Which of the following is best to sterilise heat labile compounds

(a) Dry heat (b) Autoclave
(c) Membrane filtration (d) Pasteurisation.

(v) Wash out in steady state fermentation occurs when

(a) dilution rate is less than maximum specific growth rate
(b) dilution rate is higher than the maximum specific growth rate
(c) cell concentration reaches the maximum
(d) specific growth rate is maximum.

(vi) Stationary phase is described as

(a) no further increase in the cell population after a maximum value
(b) deceleration of growth and division rate after the growth rate reaches a maximum
(c) acceleration of growth and division rate after the growth rate reaches a maximum
(d) deceleration of growth and division rate after the growth rate reaches a minimum.

(vii) The phenomenon in which substrates are used in a sequential manner is known as
(a) trans-substrate genesis (b) dialism
(c) diauxic (d) multiplicity.

(viii) If F be the volumetric flow rate, V_R be the total volume of culture in the reactor and μ be the specific growth rate, then the dilution rate, D is defined as
(a) F/V_R (b) V_R/F (c) μ/F (d) F/μ.

(ix) Which of the following is not correct for the Monod model and the Michaelis Menten Model?
(a) The Michaelis Menten Model was derived from a curve fitting exercise
(b) The Michaelis Menten model was derived from an analysis of the mechanism of microbial growth
(c) The Monod model was derived from an analysis of the mechanism of microbial growth
(d) All of the above.

(x) Bacterial growth curve is obtained by plotting
(a) number of cells versus time (b) number of spores versus time
(c) log of number of cells versus time (d) log of number of cells survived versus time.

Group – B

2.(a) How does competitive enzyme inhibition differ from non-competitive one? Explain with suitable examples.

(b) The enzyme fumarase, has the following kinetic constants:

$$\mathbf{S+E \leftrightarrows ES \rightarrow P+E,}$$

where $\mathbf{k_1 = 10^9\ M^{-1}s^{-1}}$; $\mathbf{k_{-1} = 4.4x10^4\ s^{-1}}$; $\mathbf{k_2 = 10^3 s^{-1}}$.

What is the value of the Michaelis constant for this enzyme? At an enzyme concentration of 10^{-6}M, what will be the initial rate of product formation at a substrate concentration of 10^{-3} M?

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

3.(a) Derive Michaelis-Menten Equation by quasi-steady state approach.

(b) What are the different enzyme immobilization techniques industrially used? Which one of these techniques is most commonly used and why?

6+(4+2)=12

Group – C

4. (a) How is the rate of biomass production determined for a chemostat?

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

7+5=12

5.(a) *Pseudomonas putida* with $\mu_m = 0.5\ h^{-1}$ is cultivated in a continuous culture under aerobic conditions where $D = 0.28h^{-1}$. The carbon and energy source in the feed is lactose with a concentration of $S_0 = 2$ g/l. The effluent lactose concentration is desired to be S = 0.1 g/l. If the growth rate is limited by oxygen transfer, by using the following information:

$Y^M x/s = 0.45$ gX/gS, $Y^M x/o_2 = 0.25$ gX/gO_2 and $C^* = 8$mg/l

(i) Determine the steady-state biomass concentration (X) and specific rate of oxygen consumption (qo_2).

(ii) What should be the oxygen-transfer coefficient (k_La) in order to overcome oxygen-transfer limitation (i.e. $C_L = 2$ mg/l) ?

(8+4)=12

Group – D

6.(a) Which is the most effective method for sterilization of air for use in aerobic fermentation? How is effectiveness of glass wool air filter determined?

(b) Consider the data given in the table on the temperature changes in a 10,000 lit fermenter, which includes the heat up and cool down periods. Assume initial spore concentration of 10^5/lit and vitamin concentration of 30 mg/lit. The Arhennius constant and activation energy are as follows:

<u>For spores:</u>
E= 65 Kcal/gmol
A= 10^{36} min^{-1}

<u>For vitamin:</u>
E= 10 Kcal/gmol
A= 10^4 min^{-1}.

Time (min)	0	10	20	30	40	50	60	65	90	120	140
Temperature (^oC)	30	40	54	70	95	121	121	106	75	46	32

i) What fraction of the vitamin is degraded in the heat up and sterilization period?

ii) What fraction of spores is deactivated in the heat up and sterilization period?

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

7.(a) Prove that same degree of sterilisation can be achieved from treatment at high temperature short time as from low temperature for a long time.

(b) The following experimental values were obtained for the inactivation rate constants for *Clostridium botulinum* and thiamine at 60° C and 120° C:

Temperature	Inactivation rate constant, k_d /s^{-1}	
° C	*C. botulinum*	Thiamine
60	2.42×10^{-7}	2.760×10^{-7}
120	2.42×10^{-1}	4.070×10^{-6}

Using this data, determine the time needed for a 12D reduction of thiamine activity at 135° C.

5+7=12

Group – E

8.(a) Describe the relative advantages and disadvantages of choosing the following organisms as host cell for production of protein with genetically engineered cells:
i) *E.coli* ii) Lower eukaryotic cells.

(b) What is the difference between protein secretion and protein excretion?

(c) Name two proteins which *E. coli* excretes extracellularly.

(4+4)+2+2=12

9.(a) Write two applications of mixed culture bacteria used industrially.

(b) An industrial waste with an inlet **BOD** of **800 mg/L** must be treated to reduce to exit **BOD ≤ 20mg/L**. The inlet flow rate is **400 m^3/hr**. Kinetic parameters have been estimated for waste as **μ_m = 0.20 h^{-1}**, **Ks = 50 mg/L** of **BOD, $Y^M x/s$ = 0.5 mg MLVSS/mg BOD** and **k_d = 0.005h^{-1}**. A waste treatment unit of **3200 m^3** is available. Assume a recycle ratio of **0.40** and **X_e = 0**. If **θ_c = 120h**, then find S and determine if sufficient **BOD** removal has been attained in this well mixed activated sludge process meeting these specifications. Determine X and sludge production rate of this process.

2+10=12