MANIPAL INSTITUTE OF TECHNOLOGY
MANIPAL
(A constituent unit of MAHE, Manipal)

V SEMESTER B.TECH. EXTERNAL EXAMINATIONS DEC' 2021

SUBJECT: BIOPROCESS ENGINEERING [BIO 3152]

Date of Exam: 23/12/2021 Time of Exam: 11.00 AM – 11.50 AM Max. Marks: 30

PART-A

https://forms.office.com/r/KYE47E444S

Q. No.		Marks	CO	BLT
1.	A crude extract of a cell contains 40 mg of protein/mL of solution. 20 µL of this extract was added to get a total reaction volume of 1.0 mL. The reaction resulted in the formation of 120 nmoles of product in a time of 1 minute under optimal experimental conditions. Express 'v' as nmoles/assay. a. **120 nmoles ml^{-1} min^{-1} b. 60 nmoles ml^{-1} min^{-1} c. 120 µmoles ml^{-1} min^{-1} d. 60 µmoles ml^{-1} min^{-1}	1	1	3
2.	A crude extract of a cell contains 40 mg of protein/mL of solution. 20 µL of this extract was added to get a total reaction volume of 1.0 mL. The reaction resulted in the formation of 120 nmoles of product in a time of 1 minute under optimal experimental conditions. Express 'v' as units/mL. a. **0.120 units/mL b. 0.6 units/mL c. 120 units/mL d. 60 units/mL	1	1	3
3.	A crude extract of a cell contains 40 mg of protein/mL of solution. 20 µL of this extract was added to get a total reaction volume of 1.0 mL. The reaction resulted in the formation of 120 nmoles of product in a time of 2 minute under optimal experimental conditions. Express ν as M/s. a. **1.0 x 10^{-6} M/s b. 2.0 x 10^{-6} M/s c. 0.6 x 10^{-6} M/s d. 6 x 10^{-6} M/s	1	1	3
4.	What is the initial reaction velocity at a substrate concentration of 60 mM? If K_m = 60 mM and V_{max} = 120 µM/min. Assume Michaelis-Menten kinetics is valid. a. **60 x 10^{-3} mM/min b. 6 x 10^{-3} mM/min	1	1	4

	c. 120 x 10^{-3} mM/min d. 1.20 x 10^{-3} mM/min			
5.	An enzyme was assayed at an initial substrate concentration of 6 x 10^{-5} M. In 10 minutes, half of the substrate had been consumed. K_m for this enzyme's substrate is 15 x 10^{-3} M. Calculate the reaction rate constant, 'k'. a. **0.0693 min^{-1} b. 0.115 min^{-1} c. 0.00693 min^{-1} d. 0.0115 min^{-1}	**1**	**1**	**3**
6.	An enzyme was assayed at an initial substrate concentration of 6 x 10^{-5} M. In 10 minutes, half of the substrate had been consumed. K_m for this enzyme's substrate is 15 x 10^{-3} M. Calculate V_{max}. a. **1.04 mM/min b. 0.104 mM/min c. 1.04 µM/min d. 0.104 µM/min	**1**	**1**	**4**
7.	Which of the following method of preservation can store the microorganisms for shorter period of time? a. **Growing culture with mineral oil b. Drying in vacuum c. Lyophilization d. Storage in liquid nitrogen	**1**	**2**	**1**
8.	Thermal death of microorganisms in the liquid medium follows first order kinetics. If the initial cell concentration in the fermentation medium is 10^8 cells/mL and the final acceptable contamination level is 10^{-3} cells, for how long should 1 m^3 medium be treated at temperature of 120°C (thermal deactivation rate constant = 0.23 min^{-1}) to achieve acceptable load? a. 2.0 h b. **2.83 h c. 3.0 h d. 2.5 h	**1**	**2**	**3**
9.	You are asked to develop a medium for production of an antibiotic. The antibiotic is to be made in large amounts (100000 L fermentation) and is relatively inexpensive. The host cell is a soil isolate of a fungal species, and the nutritional requirements for rapid growth are uncertain. Will you try to develop a defined or complex medium? a. Defined media b. **Undefined media	**1**	**3**	**2**
10.	Enrichment technique includes a. Either medium or incubation conditions are adjusted so as to favour the growth of the desired microorganism b. Unwanted microbes are eliminated or develop poorly since they do not find suitable growth conditions in the newly created environment c. **Both a and b d. None of the above	**1**	**2**	**2**

11.	What is the method of screening? a. To improve the microbial strain b. To improve the growth of an enzyme c. **To choose appropriate microorganisms for the desired enzyme d. To determine the optimum conditions for growth of microbes	**1**	**2**	**1**
12.	At 170°C, the specific death rate constant for X and Y microorganism has been found that 100 and 10^{10} respectively. Which one of them is less heat resistant. a. X b. **Y	**1**	**3**	**2**
13.	Logistic growth model describes which type of growth? a. Product- based growth b. Non-growth c. **Substrate-based growth d. Maintenance-based growth	**1**	**5**	**2**
14.	Maximum specific growth rate (μ_{max}) of a microorganism is calculated by taking the (ln = $\log_e$, x = biomass, t = time) a. Slope of lnx vs t of the growth cycle b. **Slope of lnx vs t during the exponential growth phase c. Slope of x vs t d. Slope of x vs t during the exponential phase of growth	**1**	**5**	**4**
15.	Yeast converts glucose to ethanol and carbon dioxide by glycosis as per the following reaction: $C_6H_{12}O_6 \rightarrow 2C_2H_5OH + 2CO_2$. Assuming complete conversion, the amount of ethanol produced (in g) from 450g of glucose is a. **230 g b. 880 g c. 120 g d. 280 g	**1**	**5**	
16.	A bacterial culture has an initial absorbance of 0.1. What is the absorbance after 1.5 h growth if the doubling time is 60 min.? a. **0.282 b. 0.345 c. 0.483 d. 0.228	**1**	**5**	
17.	*Lactobacillus* bacteria are added to well mixed and aerated medium containing 20 g/L lactose. After some time, the lactose concentration is 4 g/L and 12 g/L product is produced. Calculate the observed yield. a. 0.83 g/g b. **0.75 g/g c. 0.60 g/g d. 0.80 g/g	**1**	**5**	**5**

18.	A fermentation media is inoculated with bacteria for the production of single cell protein. If the initial substrate concentration is 25 g/L and biomass is 0.1 g/L. The substrate is then consumed and produces 9.8 g/L of biomass. What is the yield coefficient? a. **0.388 b. 0.344 c. 0.366 d. 0.318	**1**	**5**	**5**
19.	A bioreactor consists of 20,000 L of fermentation media comprised of 10 g/L of growing cells with qO_2 = 40 mmoles O_2 / (g cells h) and C_L = 2 mg O_2/L. Calculate OUR. a. 800 mmoles O_2 / (g h) b. 80 mmoles O_2 / (g h) c. **400 mmoles O_2 / (g h) d. 40 mmoles O_2 / (g h)	**1**	**4**	**3**
20.	A bioreactor consists of 20,000 L of fermentation media comprised of 10 g/L of growing cells with qO_2 = 40 mmoles O_2 / (g cells h) and C_L = 2 mg O_2/L. Calculate OTR. (Given: k_La = 180 mmol O_2/ 1 h atm, P* = 0.029 atm and P_{O2} = 0.21 atm). a. 30.05 moles O_2 / m^3 h b. **32.58 moles O_2 / m^3 h c. 31.05 moles O_2 / m^3 h d. 21.05 moles O_2 / m^3 h	**1**	**4**	**3**
21.	Enzyme is immobilized in 10 mm diameter agarose beads at a concentration of 0.02 kg protein m^{-3} gel. Ten beads are immersed in a well-mixed solution containing 4.2×10^{-3} kg m^{-3} substrate. The effective diffusivity of substrate in agarose gel is 2.1×10^{-8} m^2 s^{-1}. Kinetics of the enzyme can be approximated as first order with specific rate constant 4.11×10^4 s^{-1} per kg protein. Mass transfer effects outside the particles are negligible. Estimate the total volume of bead. a) 5.65×10^{-6} m^3 b) 5.42×10^{-6} m^3 c) 5.29×10^{-6} m^3 d)** 5.24×10^{-6} m^3	**1**	**4**	**4**
22.	Enzyme is immobilized in 10 mm diameter agarose beads at a concentration of 0.02 kg protein m^{-3} gel. Twenty beads are immersed in a well-mixed solution containing 4.2×10^{-3} kg m^{-3} substrate. The effective diffusivity of substrate in agarose gel is 2.1×10^{-8} m^2 s^{-1}. Kinetics of the enzyme can be approximated as first order with specific rate constant 4.11×10^4 s^{-1} per kg protein. Mass transfer effects outside the particles are negligible. Estimate the amount of enzyme present. a) 1.05×10^{-5} kg b) **1.05×10^{-7} kg c) 1.05×10^{-6} kg d) 1.05×10^{-8} kg	**1**	**4**	**4**
23.	Non-viable yeast cells are immobilized in alginate beads. The beads are stirred in glucose medium under anaerobic conditions. The effective diffusivity of glucose in the beads depends on cell density according to the relationship: $D_{Ac} = 5.35\text{-}6.16y_C$	**1**	**4**	**5**

	where D_{Ac} is effective diffusivity 5.25×10^{10} m^2 s^{-1} and y_C is the weight fraction of yeast in the gel. Rate of glucose uptake can be assumed to be zero order; the rate constant at a yeast density in alginate of 20 wt% is 1.0 g L^{-1} min^{-1}. For maximum reaction rate, the concentration of glucose inside the particles should remain above zero. Estimate the value of k_0, Converting k_0 to units of kg, m and s? a. **0.016 kg m^{-3} s^{-1} b. 0.006 kg m^{-3} s^{-1} c. 0.0016 kg m^{-3} s^{-1} d. 0.083 kg m^{-3} s^{-1}			
24.	To increase the overall rate of a reaction limited by internal diffusion the reaction should not _____ a) decrease the diameter D b) increase the concentration of A c) **increase the diameter D d) increase the temperature	**1**	**4**	**2**
25.	One of the following statements is not true with respect to batch sterilization a. Lower capital equipment costs. b. Lower risk if contamination c. Easier manual control d. **Under certain circumstances, the reduction of fermenter corrosion.	**1**	**3**	**2**
26.	If a pilot sterilization were carried out in 10,00,000 dm^3 vessel with a medium containing 10^{12} organisms per cm^3 requiring a probability of contamination of 1 in 1000, the sterilization criterion would be a. **55.26 b. 36.8 c. 53 d. 34.5	**1**	**3**	**4**
27.	It is required to provide a 20 m^3 fermenter with liquid media at a rate of 10 m^3/min for a fermentation lasting 200 h. The media in the fermentation plant contained approximately 400 microorganisms per m^3. Calculate the number of microorganisms present in the fermenter before sterilization. a. 96×10^7 b. **48×10^6 c. 48×10^7 d. 96×10^6	**1**	**3**	**3**
28.	When a molecule binds to an area of an enzyme that is not the active site, and changes the shape of the enzyme so that it no longer can work, this is called a. Denaturation b. Competitive inhibition c. **Noncompetitive inhibition d. Substrate inhibition	**1**	**1**	**2**
29.	Which of the following is true about Michaelis-Menten kinetics? a) K_m, the Michaelis constant, is defined as that concentration of substrate at which enzyme is working at maximum velocity	**1**	**1**	**2**

	b) **It describes single substrate enzymes c) K_m, the Michaelis constant is defined as the dissociation constant of the enzyme-substrate complex d) It assumes covalent binding occurs between enzyme and substrate			
30.	In an exponentially growing batch culture of *Lactobacillus sp.*, the cell density is 28 g/L (DCW), the specific growth rate is 0.7/h and substrate uptake rate is 24 g/L/h. The yield coefficient Y_{xs} will be a. 0.620 b. 0.600 c. 0.860 d. **0.816	**1**	**5**	**4**

CO: Course Outcome; BLOOM TAXONOMY LEVEL: 1-Remember, 2-Understand, 3-Application, 4-Analysis, 5-Evaluation, 6-Creation

Name & Signature of course coordinator | **Name & Signature of scrutinizer** | **Signature of HOD**

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SUBJECT: BIOPROCESS ENGINEERING [BIO 3152]

Date of Exam: 23/12/2021 Time of Exam: 9.20 AM – 10.45 AM Max. Marks: 20

Instructions to Candidates:

- Answer ALL the questions & missing data may be suitable assumed

PART-B

Q. No.		Marks	CO	BLT
1A.	The microbial cells are immobilized in spherical beads of agarose gel. To produce the desired enzymes the organism requires aerobic conditions. The effective diffusivity of oxygen is 1.8×10^{-8} m^2 s^{-1}. Uptake of oxygen is zero-order with intrinsic rate constant 10^{-4} mol s^{-1} m^3 particle. The concentration of oxygen at the surface of the catalyst is 10×10^{-4} kg m^{-3}. Cell growth is negligible. (a) What is the maximum particle diameter for aerobic conditions throughout the catalyst? (b) If the density of cells in the gel is reduced by a factor of four, what is the maximum particle size for aerobic conditions?	**4**	**4**	**2-4**
1B.	Yeast cells are immobilized in alginate beads. The average particle diameter is 10 mm. Rate of oxygen consumption at a bulk concentration of 10×10^{-4} kg O_2 m^{-3} is 14.2×10^{-6} kg s^{-1} m^{-3} catalyst. The effective diffusivity of oxygen in the beads is 1.88×10^{-8} m^2 s^{-1}. Assume that the oxygen concentration at the surface of the catalyst is equal to the bulk concentration, and that oxygen uptake follows First-order kinetics. a. Are internal mass-transfer effects significant? b. What reaction rate would be observed if diffusional resistance were eliminated?	**4**	**4**	**2-4**
1C.	During exponential phase in batch culture, the growth rate of a culture is proportional to the concentration of cells present. When *Bacillus sp.* are cultured in nutrient media, the concentrations of cells double in 60 min. If this rate of growth is maintained for 10 hours, what is the final concentration of the cells relative to the inoculum level?	**2**	**5**	**2**
2A.	A crude extract of a cell contains 40 mg of protein per milliliter of solution. 20 µL of this extract was added to get a total reaction volume of 1.0 mL. The reaction resulted in the formation of 120 nmoles of product in a time of 1 minute under optimal experimental conditions. Express 'v' as nmoles/assay.	**3**	**1**	**2-4**

<table>
<tr><td></td><td>a. What would ‘v’ be in the same 20 μL of the extract was assayed in a 5.0 mL total reaction volume?
b. What is the enzyme concentration in the assay mixture and on the original extract (expressed as units/mL)?
c. What is the specific activity of the preparation?</td><td></td><td></td><td></td></tr>
<tr><td>2B.</td><td>For the following data determine the k_La using gassing out method.
<table>
<tr><td>Time, s</td><td>0</td><td>40</td><td>80</td><td>120</td><td>160</td><td>200</td><td>240</td><td>280</td><td>320</td></tr>
<tr><td>DO (%)</td><td>0</td><td>38</td><td>55.8</td><td>72.4</td><td>81.3</td><td>84.7</td><td>85.5</td><td>87.8</td><td>88.2</td></tr>
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<table>
<tr><td>Time, s</td><td>360</td><td>400</td><td>440</td><td>480</td><td>520</td></tr>
<tr><td>DO (%)</td><td>89.7</td><td>94.4</td><td>91.3</td><td>90.4</td><td>91.3</td></tr>
</table></td><td>3</td><td>4</td><td>2-4</td></tr>
<tr><td>2C.</td><td>A continuous sterilizer is constructed using a 40 m length of pipe of internal diameter 10 cm. Liquid medium in the pipe is maintained at 137 ^oC using saturated steam. At this temperature, the specific death constant of the contaminating organisms is 380 h^{-1} and the density and viscosity of the medium are 1000 kg/m^3 and 1.0 cP respectively. The concentration of contaminants in the raw medium is 7 x 10^5 mL^{-1}. If sterile medium is required on the fermentation floor at a rate of 1 m^3/h, what is the frequency of contamination in the fermentation factory?</td><td>4</td><td>3</td><td>2-4</td></tr>
<tr><td colspan="5">CO: Course Outcome; BLOOM TAXONOMY LEVEL: 1-Remember, 2-Understand, 3-Application, 4-Analysis, 5-Evaluation, 6-Creation</td></tr>
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