BE. 5th Sem. Dec-2024

Exam. Code: 0909
Sub. Code: 33368

2124

B.E. (Biotechnology) Fifth Semester

BIO-511: Enzyme Engineering and Technology

Time allowed: 3 Hours **Max. Marks: 50**

NOTE: Attempt <u>five</u> questions in all, including Question No. I which is compulsory and selecting two questions from each Section. State clearly your assumptions.

x-x-x

Q. 1) Write briefly: *(1×10 =10)*

a) Define enzyme activity and specific enzyme activity?
b) Define extracellular enzyme? Give two examples.
c) Define half life of enzyme?
d) Write down the formula for the calculation of amylase activity.
e) What are the functions of protease and lipase enzyme?
f) What is the turnover number?
g) What are the cofactors? How are they useful?
h) Define effectiveness factor for immobilized enzyme?
i) Define substrate inhibition?
j) What is the Hanes –Woolf plot?

SECTION - A

Q. 2) Derive the rate expression (V) for reaction scheme given by King-Altman's method,

$$E + S \leftrightarrow ES \rightarrow E + P$$

$$E + I \leftrightarrow EI_1$$

$$EI_1 + I \leftrightarrow EI_2$$ *(10)*

Q. 3) The hydrolysis of urea by urease is only partially understood reaction and show inhibition. Data for the hydrolysis of the reaction are given in table:

Substrate concentration →	0.2 M		0.02 M	
	$\frac{1}{V}$	I	$\frac{1}{V}$	I
	0.22	0	0.68	0
	0.33	0.0012	1.02	0.0012
	0.51	0.0027	1.50	0.0022
	0.76	0.0044	1.83	0.0032
	0.88	0.0060	2.04	0.0037
	1.10	0.0080	2.72	0.0044
	1.15	0.0093	3.46	0.0060

Where V is moles/ l-min. and I is inhibitor molar concentration.

a) Determine the Michaelis-Menten constant for this reaction.
b) What type of inhibition reaction is this? Substantiate the answer.
c) Based on the answer to part (b) what is the value of K_I? *(10)*

Q. 4. a) Describe the type of enzyme inhibitions and compare V_{max} and K_m with controlled enzyme.
b) Find out degree of inhibition caused by competitive enzyme inhibition when [S] = K_m and [I] = ½ K_I
c) Explain effect of substrate and enzyme concentration on enzyme activity. *(4, 3, 3)*

P.T.O.

SECTION - B

Q. 5) Name the various methods in Block diagram. Discuss the entrapment method in details. Write advantages and disadvantages of entrapment method. How will you eliminate the enzyme leakage and diffusion problem during immobilization? *(10)*

Q. 6. a) Invertase from *Aspergillus oryzae* is immobilized in porous resin particle of diameter 1.6 mm. the effective diffusivity of sucrose in the resin is 1.3×10^{-11} m^2 s^{-1}. At a sucrose concentration of 0.85 kg m^{-3}, K_m and V_{max} for immobilized enzyme is 3.5 kg m^{-3} and 0.12 kg s^{-1} m^{-3} respectively. The observed reaction rate for free enzyme found to be 12.5 kg s^{-1} m^{-3}.

i) Calculate effectiveness factor.

ii) Determine the zero order reaction constant for immobilized invertase.

b) Derive the equation for effectiveness of an immobilized enzyme, assume that rate of substrate consumption can be expressed as zero order kinetics. *(5, 5)*

Q. 7. a) A substrate is converted to a product by the catalytic action of an enzyme. Assume that The Michaelis-Menten kinetics parameters for this reaction are:

$K_m = 0.15$ mol /L $\quad V_{max} = 4.3$ mol /L min.

i) What should be the size of steady-state CSTR to convert 95 percent to incoming substrate ($S_0 = 10$ mol/ L) with a flow rate of 10 L/ hr?

ii) What should be the size of the reactor if you employ a plug flow reactor instead of the CSTR in the part (i)?

b) The isomerisation of 5×10^{-2} mol·dm^{-1} bulk concentration of glucose to fructose is conducted at 313°K in a batch reactor using immobilised glucose isomerase. The reaction exhibits reversible Michaelis-Menten kinetics and is characterised by K_m value of 2×10^{-3} mol·dm^{-1}. The determined effectiveness factor η of 0.7 reveals an appreciable contribution of mass transport to the measured reaction rate. Calculate the substrate concentration at the solid-liquid interface under these conditions. *(4, 6)*

x-x-x