

NOTE: Attempt five questions in all, including Question No. 1 which is compulsory and selecting two questions from each Section. State clearly your assumptions.

x-x-x

1) Write briefly:

- Define enzyme activity and specific enzyme activity?
 - What are the cofactors? How are they useful?
 - Write down the formula for the calculation of amylase activity.
 - According to the Michaelis-Menten equation, what is the V/V_{max} ratio when $[S] = 3 K_m$?
 - Define Enzyme immobilization? List two advantages and disadvantages of immobilization.
- (5x2)

SECTION - A

- Derive the rate of expression (V) for different type of enzyme inhibitions.
 - Explain effect of substrate and enzyme concentration on enzyme activity.
 - 15 micrograms of an enzyme of molecular weight 30,000 working at V_{max} catalyzes the conversion of 60 micromole of substrate into product in 3 min. What is the enzyme's turnover number (in sec^{-1})?
 - The equilibrium constant for the given reaction is 5 for given reaction scheme. Suppose we have a mixture of



$$[S] = 2 \times 10^{-4} \text{ M and } [P] = 3 \times 10^{-4} \text{ M.}$$

What initial velocity will the reaction start towards equilibrium? If

$$K_m^S = 3 \times 10^{-5} \text{ M, } V_{max}^S = 2 \mu \text{ moles.lit}^{-1}\text{min.}^{-1}, V_{max}^P = 4 \mu \text{ moles.lit}^{-1}\text{min.}^{-1}. (3, 2, 2, 3)$$

- A carboxypeptidase was found to have $K_m = 2 \mu\text{M}$ and $k_{cat} = 150 \text{ s}^{-1}$ for substrate A.
 - What is the initial rate of reaction for $[A] = 5 \mu\text{M}$ and $[E_0] = 0.01 \mu\text{M}$?
 - The presence of 5 mM of a competitive inhibitor decreased the initial rate by a factor of 2. What is the value of K_i ?
 - A competing substrate B is added to part (a). Its $K_m = 10 \mu\text{M}$ and $k_{cat} = 100 \text{ s}^{-1}$. Calculate V_B/V_A .

(2, 5, 3)
- Define substrate inhibitions? Derive a rate of expression (V) for substrate inhibition kinetics and show that at maximum reaction rate of substrate concentration is

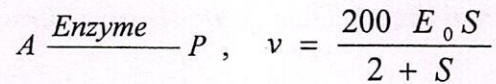
$$S_{max} = \sqrt{K_m \times K_S}$$
 - Defined Biocatalyst and what are differences between Biocatalyst and Chemical catalyst?
 - Show diagrammatically the role of enzyme in lowering the activation energy barrier.
 - After 8 minutes in batch reaction substrate ($S_0 = 1.0$ moles) is 80% converted, after 11 minutes conversion is 90%. Find V_{max} and K_m .

(3, 2, 2, 3)

(2)

SECTION - B

- 5 a) Discuss various idealized enzyme reactor systems. Discuss which you justified to be the best?
 b) Enzyme (E) catalyzes the transformation of substrate (S) to produce product (P) as follows:



If we introduce enzyme ($E_0 = 0.001$ mol/lit.) and substrate ($S_0 = 10$ mol/lit.) into a batch reactor and let the reaction proceed, find the time needed for the concentration of substrate to drop 0.025 mol/lit. (5, 5)

- 6 a) An enzyme which hydrolyzes the cellobiose to glucose, β -glycosidase is immobilized in sodium alginate gel sphere (2.5 mm in diameter). Assume that the zero order reaction occurs at every point within the sphere with $K_0 = 0.0795$ mol/s-m³ and cellobiose moves through the sphere by molecular diffusion with $D_C = 0.6 \times 10^{-5}$ cm²/s (cellobiose in gel). Calculate the effectiveness factor of the immobilized enzyme when the cellobiose concentration in bulk solution is 10 mol/m³.
- b) Immobilized lactose is used to hydrolyze lactose in dairy waste to glucose and galactose. Enzyme is immobilized in resin particles and packed into at 0.05 m³ Plug – flow column. The total effectiveness factor for the system is close to unity; K_m for the immobilized enzyme is 1.32 kg m⁻³; V_{\max} is 45 kg m⁻³ h⁻¹. The lactose concentration in the feed stream is 9.5 kg m⁻³; a substrate conversion is 98% is required. At what flow rate should the reactor be operated?
- c) 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. (3, 3, 4)
- 7) Write a critical review on “Nano flower or Metal organic frameworks (MOFs) for efficient immobilized enzyme”. (10)