

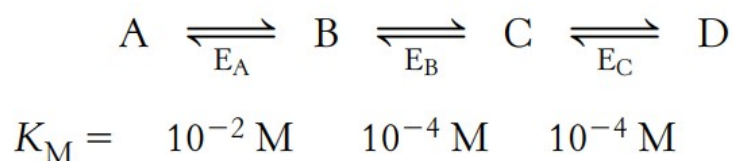
Exercises on enzyme kinetics

1 - You have isolated two versions of the same enzyme, a wild type and a mutant differing from the wild type at a single amino acid. Working carefully but expeditiously, you then establish the following kinetic characteristics of the enzymes.

	Maximum velocity	K_M
Wild type	100 $\mu\text{mol}/\text{min}$	10 mM
Mutant	1 $\mu\text{mol}/\text{min}$	0.1 mM

- (a) With the assumption that the reaction occurs in two steps in which k_{-1} is much larger than k_2 , which enzyme has the higher affinity for substrate?
- (b) What is the initial velocity of the reaction catalyzed by the wild-type enzyme when the substrate concentration is 10 mM?
- (c) Which enzyme alters the equilibrium more in the direction of product?

2 - In the conversion of A into D in the following biochemical pathway, enzymes E_A , E_B , and E_C have the K_M values indicated under each enzyme. If all of the substrates and products are present at a concentration of 10^{-4} M and the enzymes have approximately the same V_{max} , which step will be rate limiting and why?



3 - For an enzyme that follows simple Michaelis–Menten kinetics, what is the value of V_{max} if V_0 is equal to 1 mmol/minute at $10 K_M$?

4 – We obtain the following results of reaction speeds for different concentrations of substrate. The concentration of enzyme is the same in all conditions.

	Reaction speed ($\mu\text{mol/s}$)	Substrate concentration (mM)
Experiment 1	0.54	0.01
Experiment 2	4.93	0.1
Experiment 3	27.2	1
Experiment 4	45.25	5
Experiment 5	49.3	10
Experiment 6	53.76	100
Experiment 7	54.24	1000
Experiment 8	54.29	10000

- Find the value of the maximum reaction speed.
- Find the value of the Michaelis constant (K_M).
- Calculate the reaction speed when the substrate concentration is 3.5 mM.
- Explain how maximum speed can be improved.

5 - We obtain the following results of substrate produced in one minute for different concentrations of substrate. The concentration of enzyme is the same in all conditions.

	Product produced ($\mu\text{mol/min}$)	Substrate concentration (mM)
Experiment 1	4.22	0.001
Experiment 2	37.19	0.01
Experiment 3	169.03	0.1
Experiment 4	261.88	1
Experiment 5	277.1	10
Experiment 6	278.7	100
Experiment 7	278.8	1000

- Find the value of the maximum reaction speed.
- Find the value of the Michaelis constant (K_M).
- Calculate the reaction speed when the substrate concentration is 0.35 mM.

6 - We obtain the following results of reaction speeds for different concentrations of substrate. The concentration of enzyme is the same in all conditions.

	Reaction speed ($\mu\text{mol/s}$)	Substrate concentration (mM)
Experiment 1	3.8	0.1
Experiment 2	11.09	0.5
Experiment 3	14.6	1

Find the value of the maximum reaction speed and the Michaelis constant.

7 - We obtain the following results of reaction speeds for different concentrations of substrate and enzyme. These are the results we obtain:

	Reaction speed ($\mu\text{mol/s}$)	Substrate concentration (mM)	Enzyme concentration (μM)
Experiment 1	33.83	0.5	10
Experiment 2	61.02	1.0	10
Experiment 3	16.91	0.5	5

- (a) Find the value of the k_2 constant.
- (b) Find the value of the Michaelis constant.
- (c) Find the value of the maximum reaction speed for each experimental condition.