

Intro Biology Enzyme Kinetics Problems

For the reaction $A \rightleftharpoons B$ catalyzed by enzyme samples e1, e2 or e3, I obtained the following data for maximum rates for the forward reaction for the initial concentrations listed:

Enzyme #1 (e1)	Initial [A] (mM)	V (umols B produced /min)
	0	0
	1	0.2
	2	0.5
	3	0.75
	4	1
	5	1.25
	6	1.5
	7	1.73
	8	1.9
	9	1.95
	10	1.99
	15	2
	20	2
Enzyme #2 (e2)	Initial [A] (mM)	V (umols B produced /min)
	0	0
	1	0.05
	2	0.125
	3	0.1875
	4	0.25
	5	0.3125
	6	0.375
	7	0.4325
	8	0.475
	9	0.4875
	10	0.4975
	15	0.5
	20	0.5
Enzyme #3 (e3)	Initial [A] (mM)	V (umols B produced /min)
	0	0
	1	0.15
	2	0.35
	3	0.53
	4	0.68
	5	0.83
	6	0.95
	7	1.04
	8	1.13
	9	1.18
	10	1.2
	15	1.2
	20	1.2

1. Softball: Do all versions of this enzyme catalyze this particular reaction? How do you know?
2. What is the K_m for each enzyme sample?
3. Which sample has the greatest apparent $[E]$?
4. What is the difference between samples 1 and 2? Between samples 1 and 3? Samples 2 and 3?

5. Which sample will catalyze a reaction at the greatest rate if substrate is not limiting?
6. What two factors determine reaction rate for enzyme catalyzed reactions when the [A] (or any substrate) is below saturation?
7. Be able to explain the significance of affinity – we will continue to encounter this concept.

Answers:

1. Yes, since there is a reaction in each case (a measured rate of production of B).
2. K_m values: e1 = 4 mM of A, e2 = 4 mM of A and e3 = 3.5 mM of A
3. e1 since V_{max} is the greatest for the same range of [A]
4. (a) In sample 1, the [E] is 4X greater than in the e2 sample. The K_m values are the same.
 (b) There is less e3 than e1 and moreover, the K_m of e3 is lower (its affinity for A is higher). Notice that in this particular case, because there is so much less e3 than e1 that the rx rate is lower for the e3 system even though the affinity was higher. On the other hand, if the concentrations of e3 and e1 had been close, e3 would probably have a higher reaction rate at all below saturating concentrations of A.
 (c) The K_m of e2 is greater than e3 and the concentration of e3 is higher. Thus, due to both its higher concentration and greater affinity for A, the rx rate is always higher for the e3 system
5. e1 due to its high activity (and probably concentration) as seen by its high V_{MAX} .
6. K_m and "activity" (essentially due to [E]) – the former is under the control of genes and environment) it being due the interaction between the primary structure of the protein as specified by a gene and the environment and the latter is the result of (1) regulation of gene expression and (2) regulation of the protein population (by controlling the amount of active enzyme and the rate of degradation).