BL/CH401 -- Enzyme Kinetics Problems Answers

Problem 1

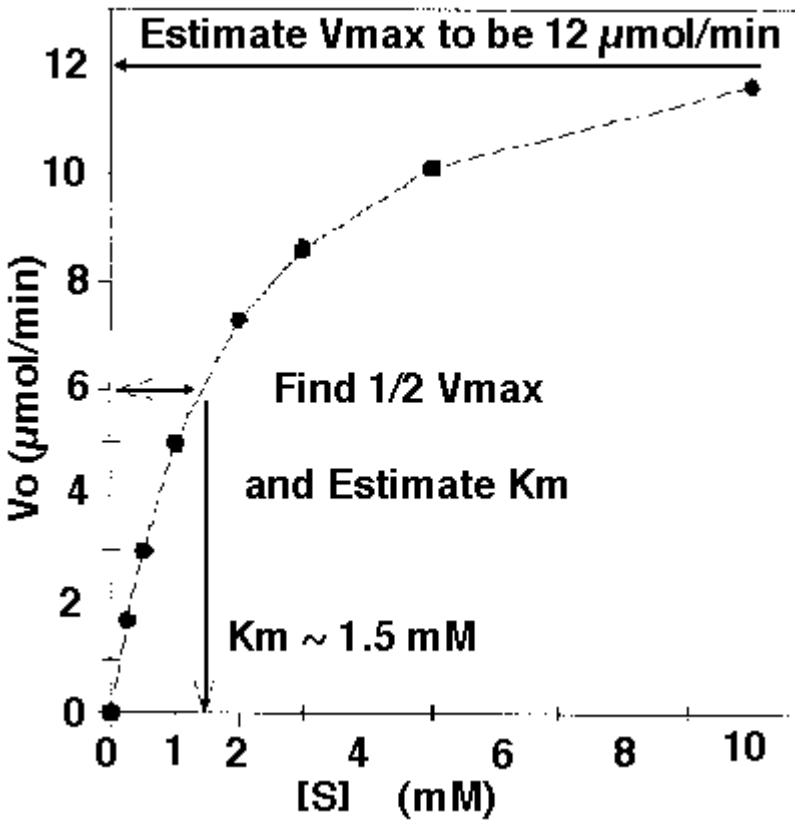

Figure 1. V vs. [S] plot

[S] mM	v_o µmol/min	1/[S] mM^{-1}	$1/v_o$ min/µmol
0.00	0.00	+Inf	+Inf
0.25	1.70	4.00	0.59
0.50	3.00	2.00	0.33
1.00	5.00	1.00	0.20
2.00	7.30	0.50	0.14
3.00	8.60	0.33	0.12
5.00	10.10	0.20	0.10
10.00	11.60	0.10	0.09

Figure 2. How to Establish Limits for the X and Y Axes on 1/Vo vs. 1[S] plot.

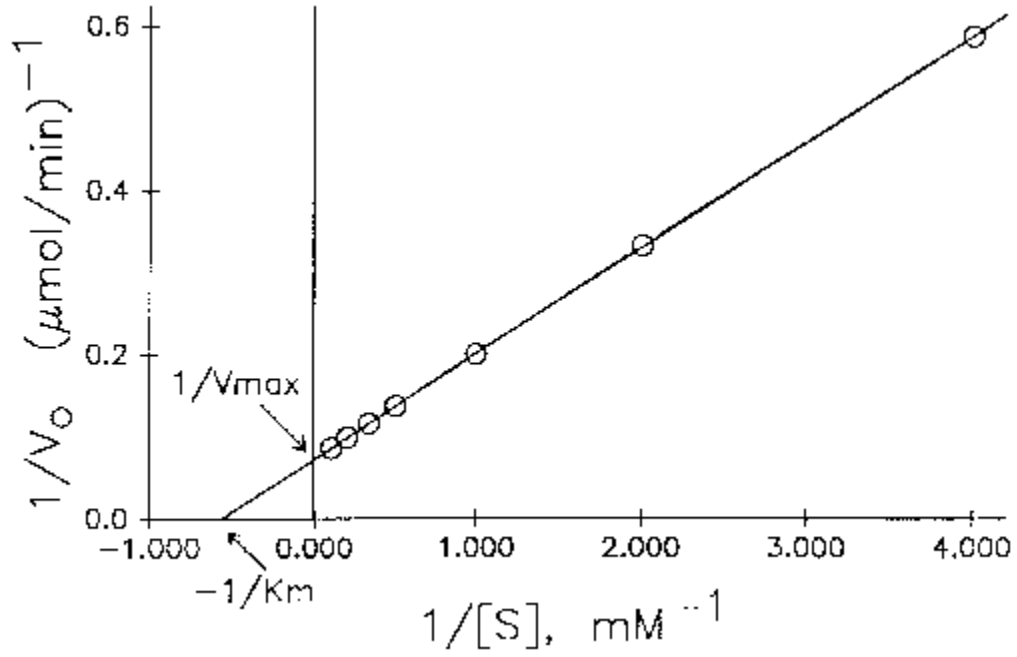

Figure 3. 1/Vo vs. 1[S] plot.

Information taken from 1/Vo vs. 1/[S] graph:

-1/Km = 0.56 so Km = 1.8 mM
1/Vmax = 0.073 so Vmax = 14 micromol/min

Don't Forget the Units for Km and Vmax
Also BE SURE to USE Correct number of Significant Figures!!

Check the Vmax and Km you found using the M-M equation:

$$v_0 = \frac{V_{max}\,[S]}{K_M + [S]}$$

Figure 4. Michaelis-Menten Equation.

Do check by plugging in Km = 1.8 mM and Vmax = 14 micromol/min;
What is Vo at [S] = 1 mM?
Vo = (14) x (1) / (1.8 + 1) = 5 micromol/min
Your answer in the check should be +/- 10% of the value given in the data set.

Problem 2

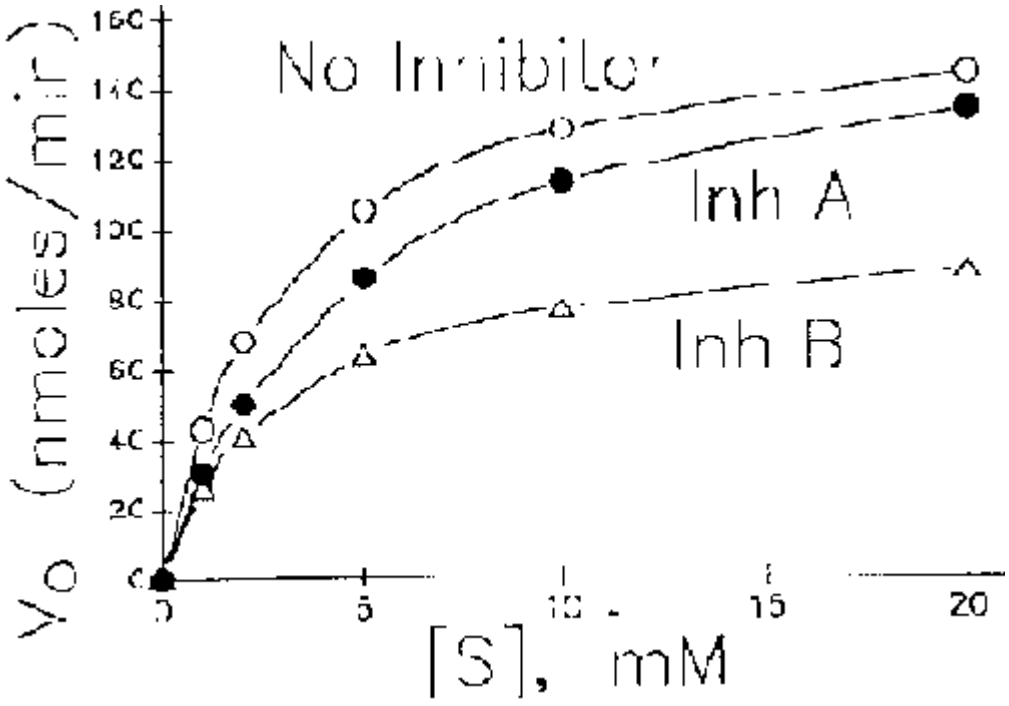

Figure 5. V vs. [S] plot

1/[S] ↓	1/vo No I	1/vo I = A	1/vo ↘ I = B
1.0000	0.0233	0.0333	0.0385
0.5000	0.0147	0.0200	0.0244
0.2000	0.0095	0.0116	0.0156
0.1000	0.0078	0.0088	0.0130
0.0500	0.0069	0.0075	0.0114

Figure 6. How to Establish Limits for the X and Y Axes on 1/Vo vs. 1[S] plot.

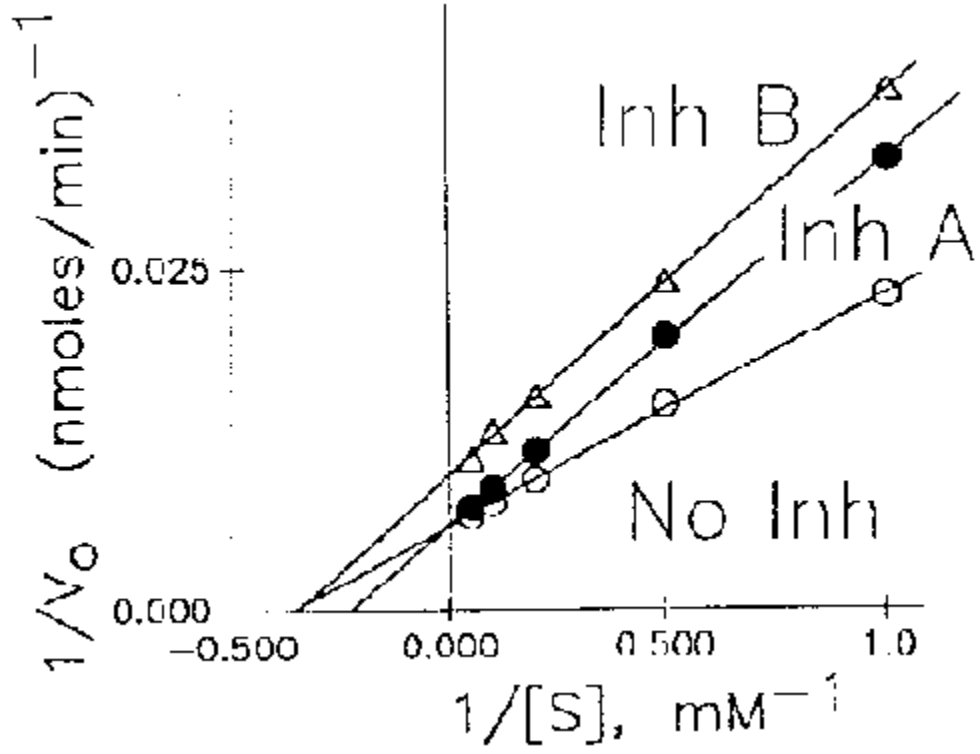

Figure 7. 1/Vo vs. 1[S] plot.

Information obtained from 1/Vo vs. 1[S] graph:

Table of Results for Kinetic Constants:

Reaction	Vmax or Vmax'	Km or Km'	Type Inhibitor
(units)	nmol/min	mM	
No Inhibitor	164	2.8	--------
Inhibitor A (5 mM)	164	4.5	Competitive Vmax' is same as Vmax
Inhibitor B (0.1 mM)	100	2.8	Non- Competitive Vmax' not equal to Vmax and Km' is same as Km

Use Summary Table to help you determine what type of Inhibitor A and B are!

A. Without Inhibitor Km = 2.8 mM and Vmax = 164 nanomol/min

Don't Forget the Units for Km and Vmax

Also BE SURE to USE Correct number of Significant Figures!!

B. In Presence of Inhibitors:

Inhibitor A = 5 mM Km' = 4.5 mM and Vmax' = 164 nmol/min;

Competitive Inhibitor; Ki = 8.3 mM

Inhibitor B = 0.1 mM Km' = 2.8 mM and Vmax' = 100 nmol/min;

Non-Competitive Inhibitor; Ki = 0.17 mM

Check the Vmax and Km you found using the M-M equation:

Do check by plugging in Km = 2.8 mM and Vmax = 164 nmol/min;

What is Vo at [S] = 5 mM?

Vo = (164) x (5) / (2.8 + 5) = 105 nmol/min

Your answer in the check should be +/- 10% of the value given in the data set.

To determine which type of Inhibitor A and B are:

For Inhibitor A, Vmax' about equal to Vmax, so it must be a Competitive Inhibitor

For Inhibitor B, Vmax' not equal to Vmax and Km' about the same as Km, so must be Non-Competitive Inhibitor.

Ki Calculations:

Choose which equation to use based on the type of Inhibitor:

Inhibitor A = Competitive

So use Km' = Km (1 + [I]/Ki)

Rearrange equation:

Ki = [I]/ ((Km'/Km) -1)

Plug [I] = 5 mM

Ki = (5 mM)/ ((4.4/2.8) -1)

Ki = 8.8 mM

Ki should have same units as I

Remember Ki can not be a negative number!

Inhibitor B = Non-Competitive

So use Vmax' = Vmax/(1 + [I]/Ki)

Rearrange equation:

Ki = [I]/((Vmax/Vmax') -1)

[I] = 0.1 mM

Ki = (0.1 mM)/((164/100)-1) = 0.16 mM

Ki should have same units as I

Remember Ki can not be a negative number!

Problem 3 (Voet & /Voet Chap 13 #4a)

Km = 3.3 mM and Vmax = 10 micromol/sec

Inhibitor 2 at 10 mM

Km' = 7.7 mM and Vmax' = 10 micromol/sec

Competitive and Ki = 7.5 mM

Inhibitor 3 at 10 mM

Km' = 3.3 mM Vmax' = 3.3 micromol/sec

Non-Competitive and Ki = 5 mM