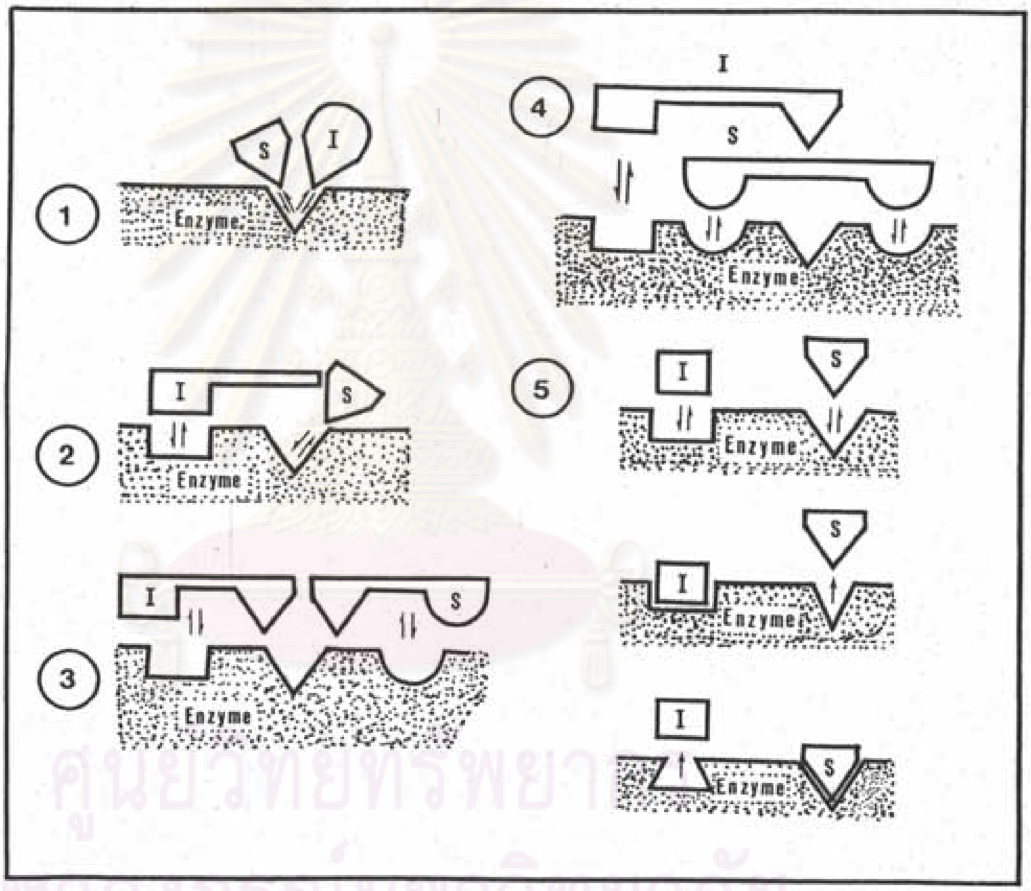
## APPENDIX I

## MODEL OF INHIBITION

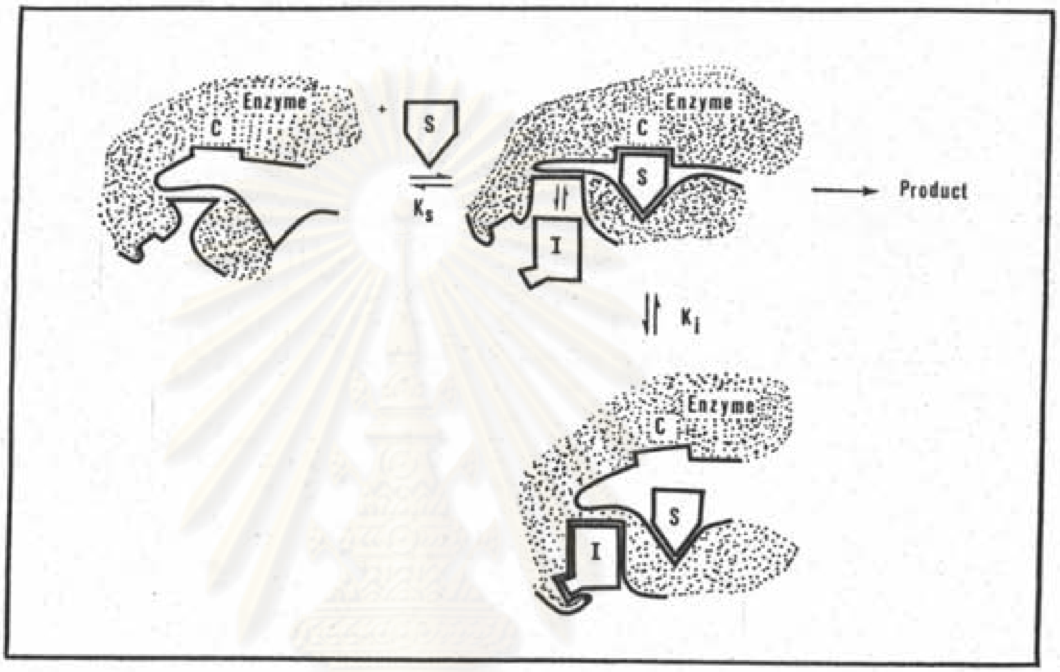
### 1. Model of Competitive Inhibition






Model of competitive inhibition; S and I are mutually exclusive. (1) Classical model S and I compete for the same binding site. I must resemble S structurally. (2) I and S are mutually exclusive because of steric hindrance. (3) I and S share a common binding group on the enzyme. (4) The binding sites for I and S are distinct, but overlapping. (5) The binding of I to a distinct inhibitor site causes a conformational change in the enzyme that distorts or masks the substrate binding site.

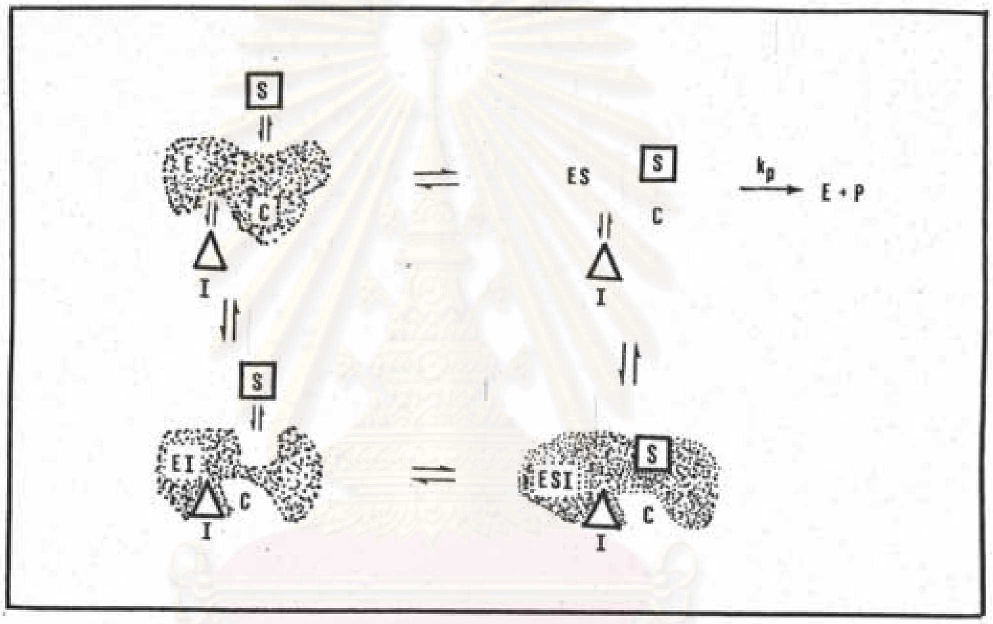
2. Model of Un-competitive Inhibition



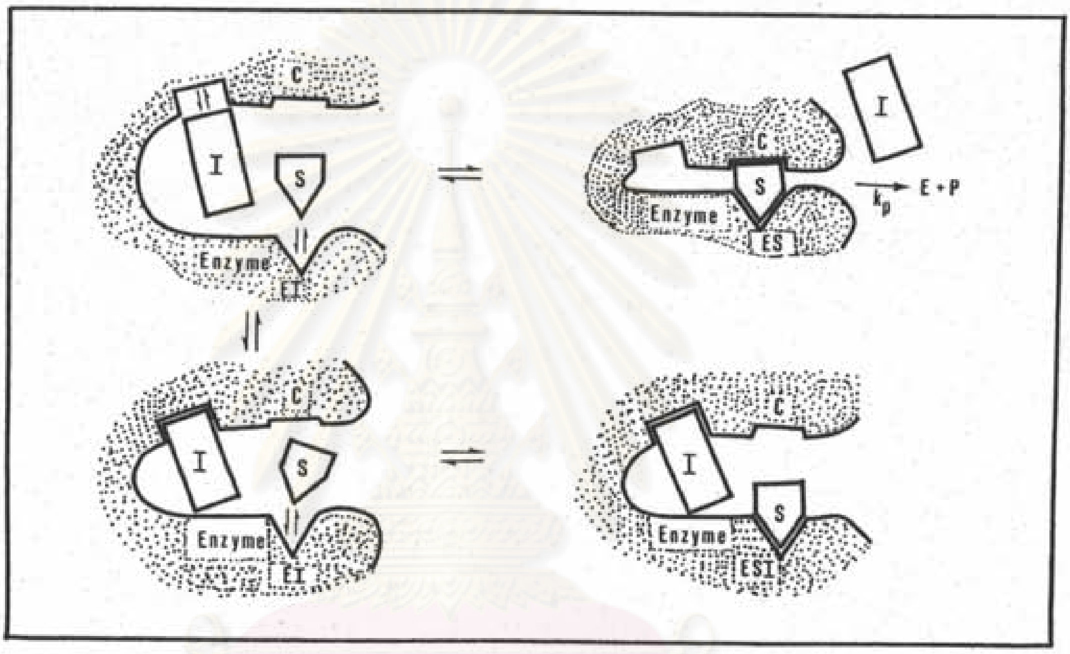

Model of un-competitive inhibition ; I binds only to ES complex. When S binds, a conformational change occurs in the enzyme which forms or unmasks the I site. The resulting ESI complex is catalytically inactive, C represents the catalytic center of the enzyme.

3. Model of Non-competitive Inhibition

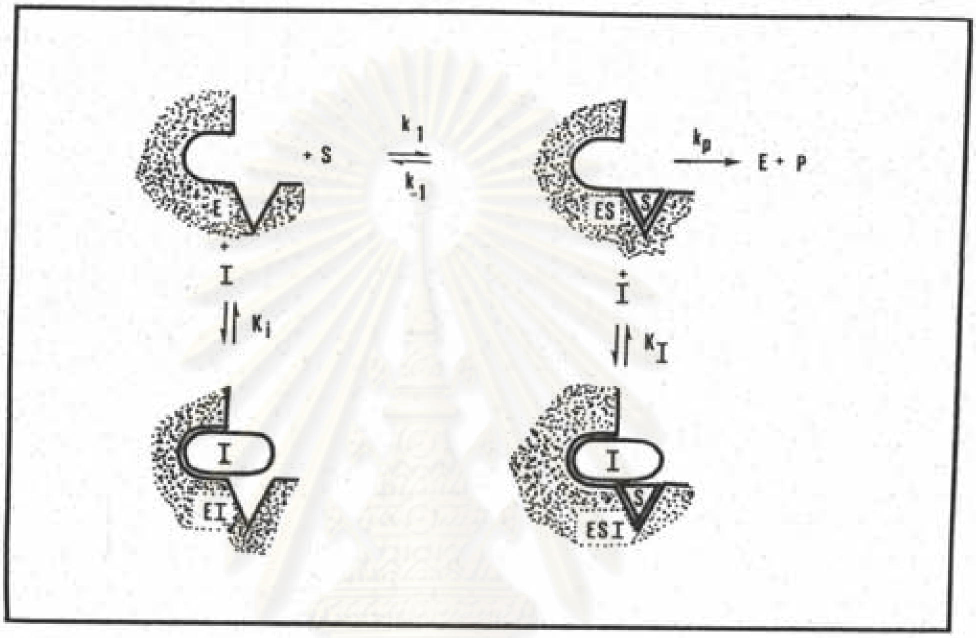
3.1 Non-competitive inhibition, S and I are not mutually exclusive but ESI is catalytically inactive. When S binds, the enzyme undergoes a conformational change which aligns the catalytic center C with the susceptible bonds of S, I interferes with the conformational change, but has no effect on S binding.



3.2 Non-competitive inhibition, In this model, I cannot bind to ES, but the properties of the system are indentical to that shown in the third model because the same four enzyme species are at equilibrium.In steady-state conditions substrate inhibition is observed.



3.3 The third model for non-competitive inhibition, I sterically hinders S binding. The velocity equation derived from steady state assumptions would be the same as that derived from rapid equilibrium assumptions.



## APPENDIX II

### A. Lineweaver-Burk Plot

The Michaelis constant, $K_m$, that is to say the equilibrium constant of the reversible combination of an enzyme with its substrate, is most conveniently determined by Lineweaver-Burk method of plotting. The great advantage of this method is that by plotting 1/V against 1/S it makes it possible to represent the Michaelis-Menten equation

$$V = \frac{V_{max}.S}{K_m + S}$$

by a straight line.

where V is the initial velocity of the reaction.

$V_{max}$ is the maximal velocity obtained at high substrate concentrations.

S is starting substrate concentration.

$K_m$ is Michaelis-Menten constant, the substrate concentration at which the reaction develops at half its maximum initial velocity.

Lineweaver-Burk calculate $K_m$ from the slope of the line and the reciprocal of the Michaelis-Menten equation,

$$\frac{1}{V} = \frac{1}{V_{max}} + \frac{K_m}{V_{max}} \times \frac{1}{S}$$

by plotting 1/V against 1/S, a straight line is obtained whose slopes are $K_m/V_{max}$ and whose intercept on the 1/V ordinate equals $1/V_{max}$ and whose intercept on the negative side of 1/S abscissa equals $-1/K_m$ .

B. <u>The Determination of Enzyme Inhibitor Constants</u>

The most widely used approach to the determination of inhibitor constant is the Lineweaver-Burk double reciprocal plot. From the slope and intercepts of these plots drawn for experiments run in the presence and absence of inhibitor, $K_m$, $K_i$ and $K_I$ may be determined. This can best be seen by presenting the reciprocal relations as they would be used, and this is done in Table II.1

Table II.1 Lineweaver-Burk Relations, Intercept and Slope Definitions

| Type | Equation | Ordinate Intercept | Slope | Abscissa Intercept |
|---|---|---|---|---|
| No inhibitor | $\frac{1}{v} = \frac{K_m}{k_2[E]_0}\left(\frac{1}{[S]}\right) + \frac{1}{k_2[E]_0}$ | $\frac{1}{k_2[E]_0}$ | $\frac{K_m}{k_2[E]_0}$ | $\frac{-1}{K_m}$ |
| a | $\frac{1}{v} = \frac{K_m\left(1 + \frac{[I]}{K_i}\right)}{k_2[E]_0}\left(\frac{1}{[S]}\right) + \frac{1}{k_2[E]_0}$ | $\frac{1}{k_2[E]_0}$ | $\frac{K_m\left(1 + \frac{[I]}{K_i}\right)}{k_2[E]_0}$ | $-\frac{1}{K_m\left(1 + \frac{[I]}{K_i}\right)}$ |
| b | $\frac{1}{v} = \frac{K_m\left(1 + \frac{[I]}{K_i}\right)}{k_2[E]_0}\left(\frac{1}{[S]}\right) + \frac{1 + \frac{[I]}{K_i}}{k_2[E]_0}$ | $\frac{1 + \frac{[I]}{K_i}}{k_2[E]_0}$ | $\frac{K_m\left(1 + \frac{[I]}{K_i}\right)}{k_2[E]_0}$ | $-\frac{1}{K_m}$ |
| c | $\frac{1}{v} = \frac{K_m}{k_2[E]_0}\left(\frac{1}{[S]}\right) + \frac{1 + \frac{[I]}{K_i}}{k_2[E]_0}$ | $\frac{1 + \frac{[I]}{K_i}}{k_2[E]_0}$ | $\frac{K_m}{k_2[E]_0}$ | $-\frac{1 + \frac{[I]}{K_i}}{K_m}$ |

a competitive

b non-competitive

c un-competitive

Enzyme inhibition may be reversible or irreversible. It is generally accepted that reversible inhibition results from noncovalent complex formation between enzyme and inhibitor. Irreversible inhibition is usually found to result from covalent bond formation between enzyme and inhibitor. Only reversible inhibition is discussed here. Three simple types of inhibition are recognized. They are termed competitive, non-competitive, and un-competitive. In addition, inhibition of an enzymatic reaction by an excess of its own substrate has been observed. These types of inhibition are generally distinguished by the effect they have on the Lineweaver-Burk plots of the enzyme-catalyzed reactions.

(a) Competitive Inhibition

The model for competitive inhibition assumes that substrate and inhibitor compete for the free enzyme; i.e. complexation of substrate, S, and inhibitor, I, to the enzyme, E, are mutually exclusive. The reactions involved are expressed by,

$$\begin{array}{lcccl} E + S & \rightleftharpoons & ES & \rightarrow & E + \text{Product} \\ K_i \updownarrow + I & & & & \\ EI & & & & \end{array}$$

where $K_i$ is the dissociation constant of the enzyme-inhibitor complex, EI. The Michaelis-Menten equation for the model of competitive inhibition becomes,

$$V = \frac{V_{max}\,[S]}{K_m\left(1+\frac{[I]}{K_i}\right) + [S]}$$

The presence of inhibitor causes $K_m$ to be multiplied by the factor $(1+[I]/K_i)$. The Lineweaver-Burk transformation becomes,

$$\frac{1}{V} = \frac{K_m}{V_{max}}\left(\frac{1}{[S]}\right)\left(1 + \frac{[I]}{K_i}\right) + \frac{1}{V_{max}} \qquad \text{(b)}$$

According to equation (b), a plot of 1/V versus 1/[S] gives a straight line with a slope of $(K_m/V_{max})(1+[I]/K_i)$. The vertical intercept at 1/[S] = 0 is $1/V_{max}$, and the intercept of the horizontal axis is $-1/K_m(1+[I]/K_i)$. Thus, the presence of a constant concentration of competitive inhibitor increases the slope of the Lineweaver-Burk plot (Fig. II.1), but has no effect on the observed value of $V_{max}$.

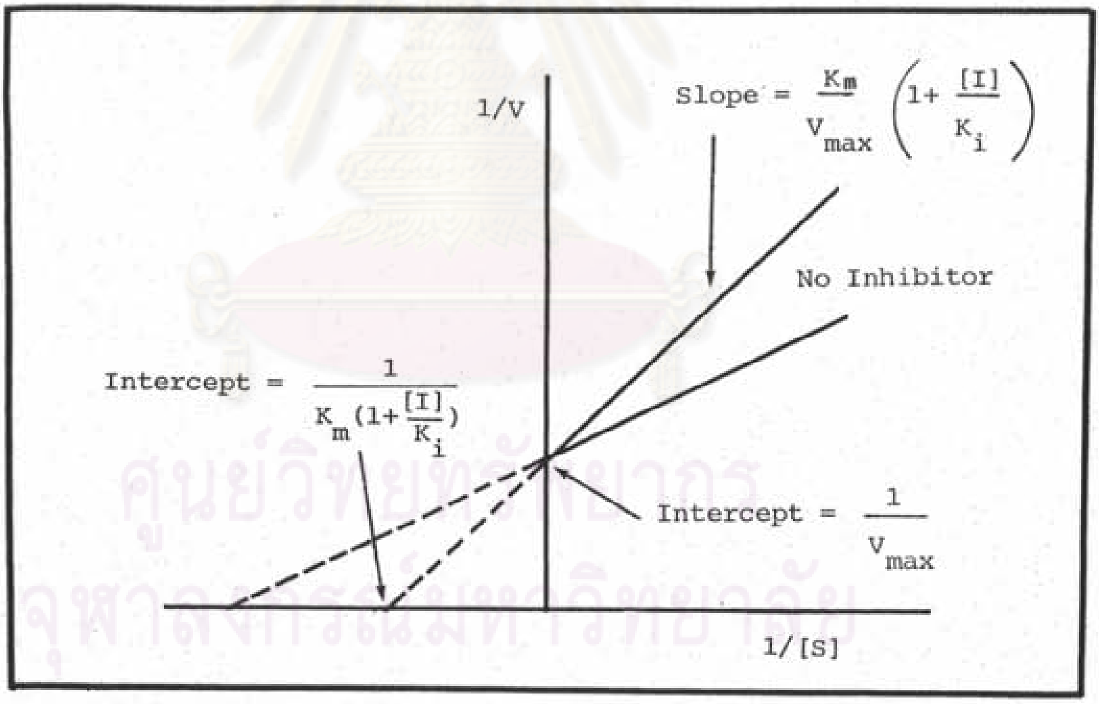



Fig. II.1 Plot of 1/V versus 1/[S] according to the method of Lineweaver-Burk

(b) Non-competitive Inhibition

A second type of inhibition which is identified on the basis of kinetic analysis is non-competitive inhibition. The simplest non-competitive inhibitors combine with the enzyme at a site other than the usual catalytic site, therefore being able to form both an EI and EIS complex neither of which can break down to yield products, so they are commonly referred as dead end complexs.

Other non-competitive inhibitors can act as partial competitive inhibitors, in that they bind in the actual substrate site and in doing so, on binding of substrate the EIS complex can either react to form normal products at a reduced rate or not react at all. All of the reactions taking place are summarized by

$$\begin{array}{ccccc} E + S & \rightleftharpoons & ES & \rightarrow & E + \text{Product} \\ K_i \updownarrow +I & & K_I \updownarrow +I & & \\ EI & \overset{+S}{\rightleftharpoons} & ESI & & \end{array}$$

For the sake of simplicity, it is assumed that $K_i$ and $K_I$ are the dissociation of inhibitor from EI and ESI. The Michaelis-Menten equation which describes the velocity of this reaction is

$$V = \frac{V_{max}[S]/(1+\frac{[I]}{K_i})}{K_m+[S]}$$

The Lineweaver-Burk transformation of this equation is

$$\frac{1}{V} = \left[\frac{K}{V_{max}}\left(\frac{1}{[S]}\right) + \frac{1}{V_{max}}\right] 1+\left(\frac{[I]}{K_i}\right)$$

A plot of 1/V versus 1/[S], according to this equation, gives a straight line which has a slope of $(K_m/V_{max})(1+[I]/K_i)$, the vertical intercept at 1/[S] = 0 is $(1/V_{max})(1+[I]/K_i)$, and the horizontal intercept at 1/V = 0 is $-1/K_m$. (Fig. II.2)

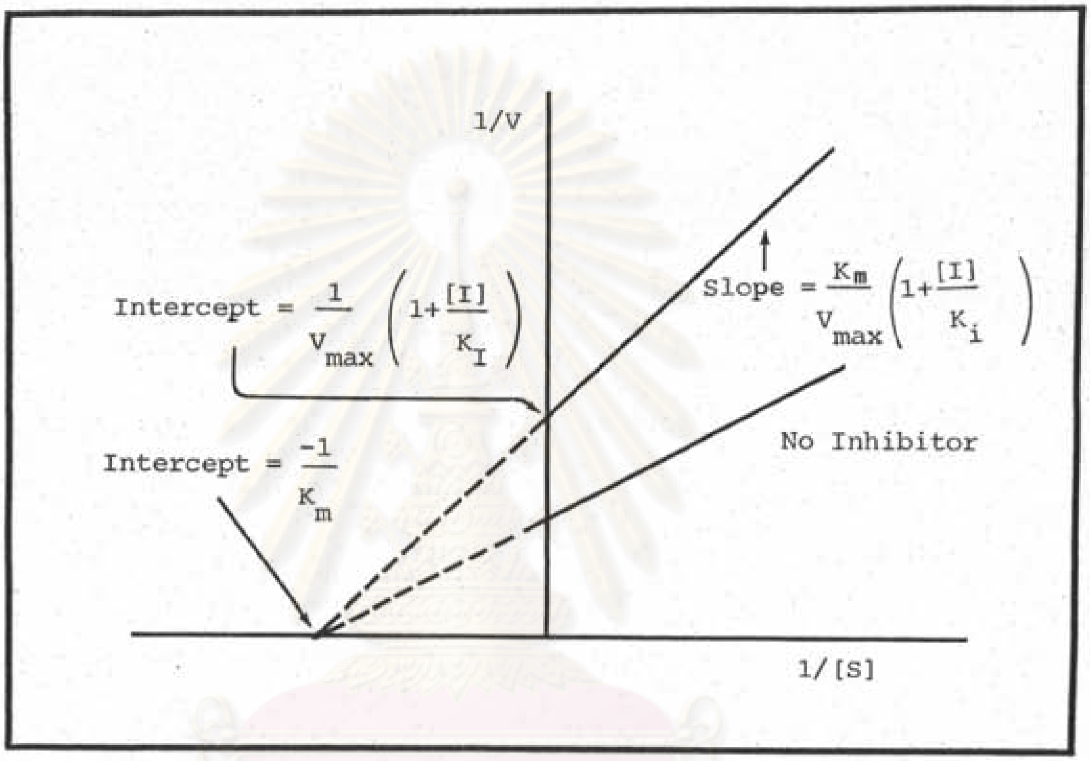



Fig. II.2 Plot of 1/V versus 1/[S] according to the method of Lineweaver-Burk

(c) <u>Un-competitive Inhibition</u>

The third type of inhibition is uncompetitive inhibition, involves an inhibitor which combines with forms of the enzymes that do not themselves combine with substrate. The ESI complex formed is a dead end complex that can only dissociate to yield I and ES with an equilibrium constant $K_i$. This type of inhibition increases the intercept on the 1/V ordinate without changing its slope. The reactions which take place are

$$E + S \rightleftharpoons ES \rightarrow E + \text{Product}$$
$$K_I \updownarrow +I$$
$$ESI$$

The overall velocity of this reaction expressed by a Michaelis-Menten type equation is

$$V = \frac{V_{max}[S]/(1+[I]/K_I)}{K_m/(1+[I]/K_I) + [S]}$$

The Lineweaver-Burk transformation of this equation is

$$\frac{1}{V} = \frac{K_m}{V_{max}}\left(\frac{1}{[S]}\right) + \frac{1}{V_{max}}\left(1+\frac{[I]}{K_I}\right)$$

A plot of 1/V versus 1/[S] for this case of un-competitive inhibition gives a straight line with a slope of $K_m/V_{max}$. This line is parallel to the line obtained in the absence of inhibitor. The verical intercept is increased to $(1/V_{max})(1+[I]/K_I)$, and the horizontal intercept is $-(1/K_m)(1+[I]/K_I)$. (Fig. II.3)

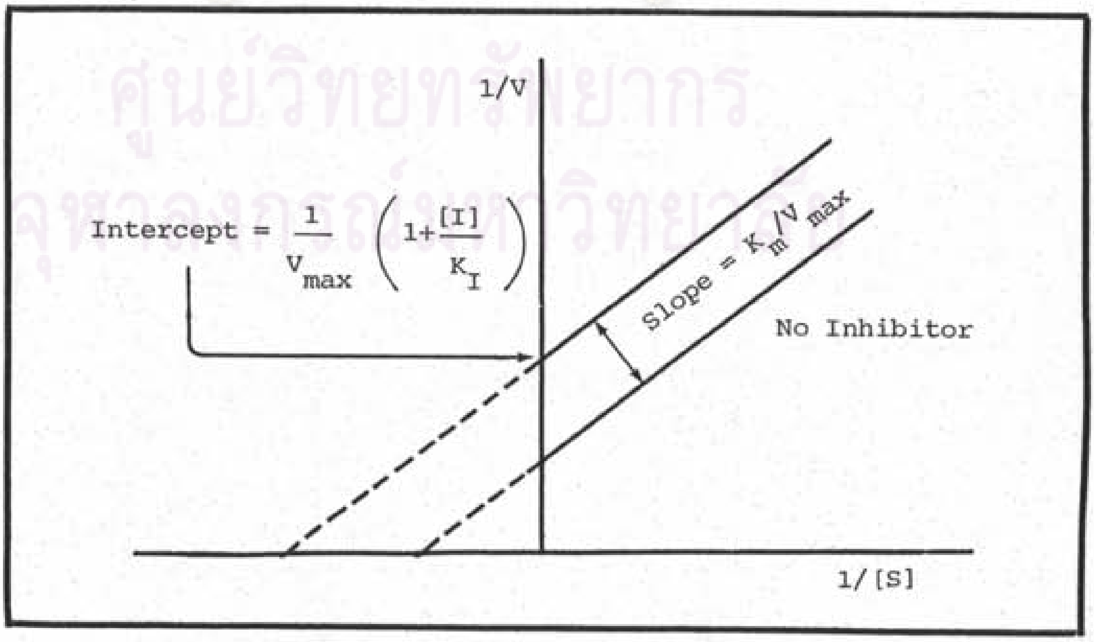



Fig. II.3 Plot of 1/V versus 1/[S] according to the method of Lineweaver-Burk

The dissociation constant of the enzyme-inhibitor complex, $K_i$, gives a quantitative measure of the strength of binding of the inhibitor to the enzyme. The parameter is evaluated easily from secondary plots of the slopes of the primary double reciprocal plot versus inhibitor concentrations. A straight line is obtained which extrapolates back somewhere to the left of the ordinate. The interception point on the [I] abscissa is the $K_i$ value of the inhibitor expressed in concentration units. A plot of these slopes versus inhibitor concentrations is linear and described for competitive and non-competitive inhibition. (Fig. II.4)

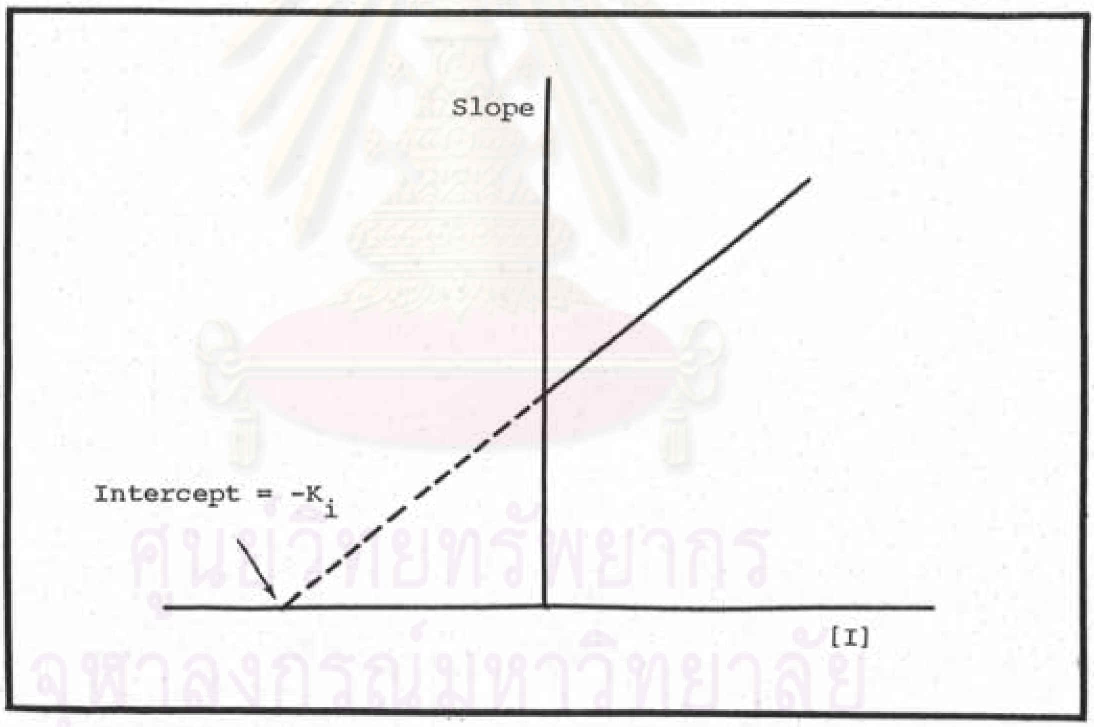



Fig. II.4 Secondary plot of inhibition data. A plot of the Slopes of Lineweaver-Burk type which shows competitive and non-competitive inhibition versus inhibitor concentration.

The dissociation constant of the enzyme-substrate-Inhibitor complex, $K_I$, gives a quantitative measure of the strength of binding of the inhibitor to the enzyme-substrate complex. This parameter is evaluated easily from secondary plots of the intercepts of the primary double reciprocal plots versus inhibitor concentration. A straight line is obtained which is extrapolated back to the [I] abscissa, which gives the $K_I$ value expressed in concentration units. A plot of the intercepts versus the inhibitor concentrations is linear and described for non-competitive and un-competitive inhibitions. (Fig. II.5)

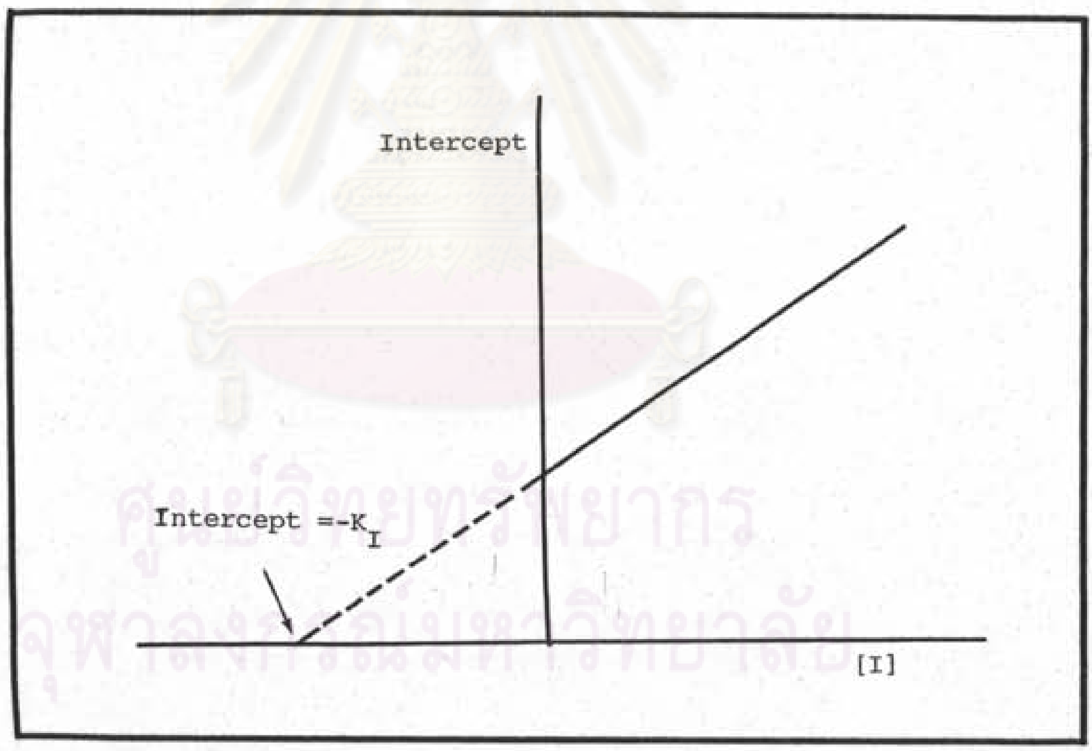





Fig. II.5 Secondary plot of inhibition data. A plot of the intercepts of Lineweaver-Burk type which shows uncompetitive and non-competitive inhibition versus inhibitor concentration.

APPENDIX III

DATA FOR THE DETERMINATION OF TYPES OF INHIBITION

Table III.1 Substrate solutions for the determination of the types of inhibition.

| [BAN] ($\times 10^{-3}$M) | Amount of BAN (µl) | $H_2O$ (µl) | HEPES (µl) | DMSO (µl) | Enzyme (µl) | Amount of Inhibitor (µl) |
|---|---|---|---|---|---|---|
| 0.1 | 150 | 2x525 | 2x500 | 300 | 500 | 0 |
| 0.08 | 120 | 2x525 | 2x500 | 330 | 500 | 0 |
| 0.06 | 90 | 2x525 | 2x500 | 360 | 500 | 0 |
| 0.04 | 60 | 2x525 | 2x500 | 390 | 500 | 0 |
| 0.02 | 30 | 2x525 | 2x500 | 420 | 500 | 0 |
| 0.1 | 150 | 2x525 | 2x500 | 150 | 500 | 150 |
| 0.08 | 120 | 2x525 | 2x500 | 180 | 500 | 150 |
| 0.06 | 90 | 2x525 | 2x500 | 210 | 500 | 150 |
| 0.04 | 60 | 2x525 | 2x500 | 240 | 500 | 150 |
| 0.02 | 30 | 2x525 | 2x500 | 270 | 500 | 150 |
| 0.1 | 150 | 2x525 | 2x500 | 100 | 500 | 200 |
| 0.08 | 120 | 2x525 | 2x500 | 130 | 500 | 200 |
| 0.06 | 90 | 2x525 | 2x500 | 160 | 500 | 200 |
| 0.04 | 60 | 2x525 | 2x500 | 190 | 500 | 200 |
| 0.02 | 30 | 2x525 | 2x500 | 210 | 500 | 200 |

Table III.2 Lineweaver-Burk data of compound 1 (trypsin)

| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
|---|---|---|---|---|
| Inhibitor = 0 | | | | |
| 0.10 | 0.146 | 6.85 | 10.00 | Slope ($K_m/V_{max}$) = 0.0762 |
| 0.08 | 0.142 | 7.04 | 12.50 | y = 6.0761 |
| 0.06 | 0.136 | 7.35 | 16.67 | x = -79.7438 |
| 0.04 | 0.126 | 7.94 | 25.00 | Correlation coefficient = 0.0998 |
| 0.02 | 0.101 | 9.90 | 50.00 | |
| Inhibitor = 150 $\mu$l. | | | | |
| 0.10 | 0.123 | 8.13 | 10.00 | Slope ($K_m/V_{max}$) = 0.0769 |
| 0.08 | 0.119 | 8.40 | 12.50 | y = 7.4051 |
| 0.06 | 0.115 | 8.69 | 16.67 | x = -96.3519 |
| 0.04 | 0.107 | 9.34 | 25.00 | Correlation coefficient = 0.9997 |
| 0.02 | 0.089 | 11.24 | 50.00 | |
| Inhibitor = 200 $\mu$l. | | | | |
| 0.10 | 0.101 | 9.90 | 10.00 | Slope ($K_m/V_{max}$) = 0.0770 |
| 0.08 | 0.099 | 10.10 | 12.50 | y = 9.1215 |
| 0.06 | 0.096 | 10.42 | 16.67 | x = -118.4405 |
| 0.04 | 0.091 | 10.99 | 25.00 | Correlation coefficient = 0.9997 |
| 0.02 | 0.077 | 12.99 | 50.00 | |

Table III.3 Lineweaver-Burk data of compound 2 (trypsin)

| $BAN \times 10^{-3}$ | v | 1/v | $1/BAN \times 10^{3}$ | Regression Analysis |
|---|---|---|---|---|
| Inhibitor = 0 | | | | |
| 0.10 | 0.175 | 5.71 | 10.00 | Slope ($K_m/V_{max}$) = 0.0803 |
| 0.08 | 0.169 | 5.92 | 12.50 | y = 4.9055 |
| 0.06 | 0.160 | 6.25 | 16.67 | x = -61.0600 |
| 0.04 | 0.145 | 6.89 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.112 | 8.93 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.152 | 6.58 | 10.00 | Slope ($K_m/V_{max}$) = 0.0831 |
| 0.08 | 0.147 | 6.80 | 12.50 | y = 5.7385 |
| 0.06 | 0.141 | 7.09 | 16.67 | x = -69.0533 |
| 0.04 | 0.128 | 7.81 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.101 | 9.90 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.139 | 7.19 | 10.00 | Slope ($K_m/V_{max}$) = 0.0800 |
| 0.08 | 0.135 | 7.41 | 12.50 | y = 6.4318 |
| 0.06 | 0.128 | 7.81 | 16.67 | x = -80.4188 |
| 0.04 | 0.118 | 8.47 | 25.00 | Correlation coefficient = 0.9995 |
| 0.02 | 0.096 | 10.41 | 50.00 | |

Table III.4 Lineweaver-Burk data of compound 3 (trypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
| 0.10 | 0.133 | 7.52 | 10.00 | Slope $(K_m/V_{max})$ = 0.1728 |
| 0.08 | 0.126 | 7.94 | 12.50 | y = 5.6353 |
| 0.06 | 0.116 | 8.71 | 16.67 | x = -32.6195 |
| 0.04 | 0.101 | 9.90 | 25.00 | Correlation coefficient = 0.9975 |
| 0.02 | 0.070 | 14.28 | 50.00 | |
| Inhibitor = 150 $\mu$l. | | | | |
| 0.10 | 0.111 | 9.01 | 10.00 | Slope $(K_m/V_{max})$ = 0.1993 |
| 0.08 | 0.105 | 9.52 | 12.50 | y = 6.9957 |
| 0.06 | 0.098 | 10.20 | 16.67 | x = -35.1050 |
| 0.04 | 0.083 | 12.05 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.059 | 16.95 | 50.00 | |
| Inhibitor = 200 $\mu$l. | | | | |
| 0.10 | 0.101 | 9.90 | 10.00 | Slope $(K_m/V_{max})$ = 0.2138 |
| 0.08 | 0.095 | 10.53 | 12.50 | y = 7.8389 |
| 0.06 | 0.087 | 11.49 | 16.67 | x = -36.6702 |
| 0.04 | 0.075 | 13.16 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.054 | 18.52 | 50.00 | |

Table III.5 Lineweaver-Burk data of compound 4 (trypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
| 0.10 | 0.082 | 12.20 | 10.00 | Slope ($K_m/V_{max}$) = 0.2919 |
| 0.08 | 0.078 | 12.82 | 12.50 | y = 9.1058 |
| 0.06 | 0.072 | 13.89 | 16.67 | x = -31.1995 |
| 0.04 | 0.062 | 16.13 | 25.00 | Correlation coefficient = 0.9993 |
| 0.02 | 0.042 | 23.18 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.053 | 18.87 | 10.00 | Slope ($K_m/V_{max}$) = 0.3633 |
| 0.08 | 0.051 | 19.61 | 12.50 | y = 15.2000 |
| 0.06 | 0.047 | 21.28 | 16.67 | x = -41.8370 |
| 0.04 | 0.041 | 24.39 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.030 | 33.33 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.047 | 21.28 | 10.00 | Slope ($K_m/V_{max}$) = 0.3633 |
| 0.08 | 0.045 | 22.22 | 12.50 | y = 17.5705 |
| 0.06 | 0.041 | 24.39 | 16.67 | x = -44.7031 |
| 0.04 | 0.036 | 27.77 | 25.00 | Correlation coefficient = 0.9989 |
| 0.02 | 0.027 | 37.04 | 50.00 | |

Table III.6 Lineweaver-Burk data of compound 5 (trypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
| 0.10 | 0.231 | 4.33 | 10.00 | Slope ($K_m/V_{max}$) = 0.0827 |
| 0.08 | 0.220 | 4.54 | 12.50 | y = 3.4877 |
| 0.06 | 0.207 | 4.83 | 16.67 | x = -42.1550 |
| 0.04 | 0.180 | 5.55 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.131 | 7.63 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.186 | 5.38 | 10.00 | Slope ($K_m/V_{max}$) = 0.0984 |
| 0.08 | 0.178 | 5.62 | 12.50 | y = 4.3498 |
| 0.06 | 0.165 | 6.06 | 16.67 | x = -44.2178 |
| 0.04 | 0.152 | 6.58 | 25.00 | Correlation coefficient = 0.9968 |
| 0.02 | 0.107 | 9.34 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.115 | 8.70 | 10.00 | Slope ($K_m/V_{max}$) = 0.1388 |
| 0.08 | 0.110 | 9.09 | 12.50 | y = 7.3609 |
| 0.06 | 0.103 | 9.71 | 16.67 | x = -53.0369 |
| 0.04 | 0.092 | 10.87 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.070 | 14.28 | 50.00 | |

Table III.7 Lineweaver-Burk data of compound 6 (trysin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| BANx10$^{-3}$ | V | 1/V | 1/BANx10$^{3}$ | Regression Analysis |
| 0.10 | 0.117 | 8.55 | 10.00 | Slope ($K_m/V_{max}$) = 0.1155 |
| 0.08 | 0.113 | 8.85 | 12.50 | y = 7.3877 |
| 0.06 | 0.108 | 9.26 | 16.67 | x = -63.9399 |
| 0.04 | 0.097 | 10.31 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.076 | 13.16 | 50.00 | |
| Inhibitor = 150 μl. | | | | |
| 0.10 | 0.089 | 11.24 | 10.00 | Slope ($K_m/V_{max}$) = 0.1423 |
| 0.08 | 0.086 | 11.63 | 12.50 | y = 9.8213 |
| 0.06 | 0.082 | 12.20 | 16.67 | x = 69.0316 |
| 0.04 | 0.075 | 13.33 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.059 | 16.95 | 50.00 | |
| Inhibitor = 200 μl. | | | | |
| 0.10 | 0.078 | 12.82 | 10.00 | Slope ($K_m/V_{max}$) = 0.1602 |
| 0.08 | 0.076 | 13.16 | 12.50 | y = 11.1457 |
| 0.06 | 0.072 | 13.89 | 16.67 | x = -69.5677 |
| 0.04 | 0.067 | 14.92 | 25.00 | Correlation coefficient = 0.9987 |
| 0.02 | 0.052 | 19.23 | 50.00 | |

Table III.8 Lineweaver-Burk data of compound 7 (trypsin)

| $BAN \times 10^{-3}$ | V | 1/V | $1/BAN \times 10^{3}$ | Regression Analysis |
|---|---|---|---|---|
| Inhibitor = 0 | | | | |
| 0.10 | 0.175 | 5.71 | 10.00 | Slope ($K_m/V_{max}$) = 0.1871 |
| 0.08 | 0.162 | 6.17 | 12.50 | y = 3.7891 |
| 0.06 | 0.147 | 6.80 | 16.67 | x = -20.2488 |
| 0.04 | 0.118 | 8.47 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.076 | 13.16 | 50.00 | |
| Inhibitor = 150 $\mu$l. | | | | |
| 0.10 | 0.122 | 8.20 | 10.00 | Slope ($K_m/V_{max}$) = 0.2426 |
| 0.08 | 0.115 | 8.69 | 12.50 | y = 5.7236 |
| 0.06 | 0.102 | 9.80 | 16.67 | x = -23.5973 |
| 0.04 | 0.085 | 11.76 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.056 | 17.86 | 50.00 | |
| Inhibitor = 200 $\mu$l. | | | | |
| 0.10 | 0.105 | 9.52 | 10.00 | Slope ($K_m/V_{max}$) = 0.2726 |
| 0.08 | 0.098 | 10.20 | 12.50 | y = 6.7509 |
| 0.06 | 0.089 | 11.24 | 16.67 | x = -24.7625 |
| 0.04 | 0.074 | 13.51 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.049 | 20.41 | 50.00 | |

Table III.9 Lineweaver-Burk data of compound 8 (trypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
| 0.10 | 0.176 | 5.68 | 10.00 | Slope ($K_m/V_{max}$) = 0.0995 |
| 0.08 | 0.167 | 5.99 | 12.50 | y = 4.6934 |
| 0.06 | 0.156 | 6.41 | 16.67 | x = -47.1564 |
| 0.04 | 0.142 | 7.04 | 25.00 | Correlation coefficient = 0.9987 |
| 0.02 | 0.103 | 9.71 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.123 | 8.13 | 10.00 | Slope ($K_m/V_{max}$) = 0.1236 |
| 0.08 | 0.119 | 8.40 | 12.50 | y = 6.9686 |
| 0.06 | 0.111 | 9.01 | 16.67 | x = -52.5592 |
| 0.04 | 0.090 | 11.11 | 25.00 | Correlation coefficient = 0.9775 |
| 0.02 | 0.075 | 13.33 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.107 | 9.34 | 10.00 | Slope ($K_m/V_{max}$) = 0.1454 |
| 0.08 | 0.103 | 9.71 | 12.50 | y = 7.9078 |
| 0.06 | 0.097 | 10.31 | 16.67 | x = -54.3850 |
| 0.04 | 0.086 | 11.63 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.066 | 15.15 | 50.00 | |

Table III.10 Lineweaver-Burk data of compound 9 (trypsin)

| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
|---|---|---|---|---|
| Inhibitor = 0 | | | | |
| 0.10 | 0.122 | 8.20 | 10.00 | Slope $(K_m/V_{max})$ = 0.1522 |
| 0.08 | 0.117 | 8.55 | 12.50 | y = 6.6266 |
| 0.06 | 0.109 | 9.17 | 16.67 | x = -43.5385 |
| 0.04 | 0.097 | 10.31 | 25.00 | Correlation coefficient = 0.9996 |
| 0.02 | 0.070 | 14.28 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.119 | 8.40 | 10.00 | Slope $(K_m/V_{max})$ = 0.1742 |
| 0.08 | 0.113 | 8.85 | 12.50 | y = 6.6937 |
| 0.06 | 0.104 | 9.62 | 16.67 | x = -38.4195 |
| 0.04 | 0.090 | 11.11 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.065 | 15.38 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.114 | 8.77 | 10.00 | Slope $(K_m/V_{max})$ = 0.2350 |
| 0.08 | 0.107 | 9.34 | 12.50 | y = 6.4431 |
| 0.06 | 0.096 | 10.42 | 16.67 | x = -27.4130 |
| 0.04 | 0.081 | 12.34 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.055 | 18.18 | 50.00 | |

Table III.11 Lineweaver-Burk data of compound 10 (trypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| BANx$10^{-3}$ | V | 1/V | 1/BANx$10^{3}$ | Regression Analysis |
| 0.10 | 0.145 | 6.89 | 10.00 | Slope ($K_m/V_{max}$) = 0.1660 |
| 0.08 | 0.137 | 7.30 | 12.50 | y = 5.2242 |
| 0.06 | 0.126 | 7.94 | 16.67 | x = -31.4767 |
| 0.04 | 0.106 | 9.43 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.074 | 13.51 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.107 | 9.34 | 10.00 | Slope ($K_m/V_{max}$) = 0.2203 |
| 0.08 | 0.101 | 9.90 | 12.50 | y = 7.1248 |
| 0.06 | 0.092 | 10.86 | 16.67 | x = -32.3357 |
| 0.04 | 0.080 | 12.50 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.055 | 18.18 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.081 | 12.34 | 10.00 | Slope ($K_m/V_{max}$) = 0.2844 |
| 0.08 | 0.076 | 13.16 | 12.50 | y = 9.5999 |
| 0.06 | 0.069 | 14.49 | 16.67 | x = -33.7542 |
| 0.04 | 0.060 | 16.67 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.042 | 23.81 | 50.00 | |

Table III.12 Lineweaver-Burk data of compound 11 (trypsin)

| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
|---|---|---|---|---|
| Inhibitor = 0 | | | | |
| 0.10 | 0.127 | 7.87 | 10.00 | Slope ($K_m/V_{max}$) = 0.1033 |
| 0.08 | 0.123 | 8.13 | 12.50 | y = 6.8911 |
| 0.06 | 0.114 | 8.77 | 16.67 | x = -66.7043 |
| 0.04 | 0.106 | 9.43 | 25.00 | Correlation coefficient = 0.9986 |
| 0.02 | 0.083 | 12.05 | 50.00 | |
| Inhibitor = 150 $\mu$l. | | | | |
| 0.10 | 0.122 | 8.20 | 10.00 | Slope ($K_m/V_{max}$) = 0.1340 |
| 0.08 | 0.119 | 8.40 | 12.50 | y = 6.8194 |
| 0.06 | 0.110 | 9.09 | 16.67 | x = -50.8773 |
| 0.04 | 0.098 | 10.20 | 25.00 | Correlation coefficient = 0.9997 |
| 0.02 | 0.074 | 13.51 | 50.00 | |
| Inhibitor = 200 $\mu$l. | | | | |
| 0.10 | 0.112 | 8.93 | 10.00 | Slope ($K_m/V_{max}$) = 0.2077 |
| 0.08 | 0.106 | 9.43 | 12.50 | y = 6.8200 |
| 0.06 | 0.097 | 10.31 | 16.67 | x = -32.8402 |
| 0.04 | 0.084 | 11.90 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.058 | 17.24 | 50.00 | |

Table III.13 Lineweaver-Burk data of compound 12 (trypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
| 0.10 | 0.117 | 8.55 | 10.00 | Slope ($K_m/V_{max}$) = 0.1341 |
| 0.08 | 0.113 | 8.85 | 12.50 | y = 7.2106 |
| 0.06 | 0.106 | 9.43 | 16.67 | x = -53.7811 |
| 0.04 | 0.094 | 10.64 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.072 | 13.89 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.110 | 9.09 | 10.00 | Slope ($K_m/V_{max}$) = 0.1888 |
| 0.08 | 0.104 | 9.62 | 12.50 | y = 7.2589 |
| 0.06 | 0.096 | 10.42 | 16.67 | x = -38.4469 |
| 0.04 | 0.083 | 12.05 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.060 | 16.67 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.103 | 9.71 | 10.00 | Slope ($K_m/V_{max}$) = 0.2484 |
| 0.08 | 0.096 | 10.42 | 12.50 | y = 7.4046 |
| 0.06 | 0.088 | 11.36 | 16.67 | x = -29.8124 |
| 0.04 | 0.070 | 14.28 | 25.00 | Correlation coefficient = 0.9957 |
| 0.02 | 0.051 | 19.61 | 50.00 | |

Table III.14 Lineweaver-Burk data of compound 13 (trypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| BANx$10^{-3}$ | V | 1/V | 1/BANx$10^{3}$ | Regression Analysis |
| 0.10 | 0.119 | 8.40 | 10.00 | Slope ($K_m/V_{max}$) = 0.1936 |
| 0.08 | 0.113 | 8.85 | 12.50 | y = 6.4698 |
| 0.06 | 0.103 | 9.71 | 16.67 | x = -33.4223 |
| 0.04 | 0.088 | 11.36 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.062 | 16.13 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.109 | 9.17 | 10.00 | Slope ($K_m/V_{max}$) = 0.2616 |
| 0.08 | 0.102 | 9.80 | 12.50 | y = 6.5483 |
| 0.06 | 0.092 | 10.87 | 16.67 | x = -25.0303 |
| 0.04 | 0.076 | 13.16 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.051 | 19.61 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.100 | 10.00 | 10.00 | Slope ($K_m/V_{max}$) = 0.3609 |
| 0.08 | 0.091 | 10.99 | 12.50 | y = 6.3185 |
| 0.06 | 0.083 | 12.04 | 16.67 | x = -17.5059 |
| 0.04 | 0.065 | 15.38 | 25.00 | Correlation coefficient = 0.9996 |
| 0.02 | 0.041 | 24.39 | 50.00 | |

Table III.15 Lineweaver-Burk data of compound 14 (trypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
| 0.10 | 0.106 | 9.43 | 10.00 | Slope $(K_m/V_{max})$ = 0.1667 |
| 0.08 | 0.101 | 9.90 | 12.50 | y = 7.8242 |
| 0.06 | 0.094 | 10.64 | 16.67 | x = -46.9427 |
| 0.04 | 0.083 | 12.05 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.062 | 16.13 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.102 | 9.80 | 10.00 | Slope $(K_m/V_{max})$ = 0.2244 |
| 0.08 | 0.095 | 10.53 | 12.50 | y = 7.6807 |
| 0.06 | 0.087 | 11.49 | 16.67 | x = -34.2318 |
| 0.04 | 0.075 | 13.33 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.053 | 18.87 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.095 | 10.53 | 10.00 | Slope $(K_m/V_{max})$ = 0.2694 |
| 0.08 | 0.089 | 11.24 | 12.50 | y = 7.8390 |
| 0.06 | 0.082 | 12.20 | 16.67 | x = -29.1002 |
| 0.04 | 0.068 | 14.70 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.047 | 21.28 | 50.00 | |

Table III.16 Lineweaver-Burk data of compound 15 (trypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| BANx$10^{-3}$ | V | 1/V | 1/BANx$10^{3}$ | Regression Analysis |
| 0.10 | 0.153 | 6.53 | 10.00 | Slope ($K_m/V_{max}$) = 0.1339 |
| 0.08 | 0.146 | 6.85 | 12.50 | y = 5.2134 |
| 0.06 | 0.133 | 7.52 | 16.67 | x = -38.9459 |
| 0.04 | 0.117 | 8.55 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.084 | 11.90 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.144 | 6.94 | 10.00 | Slope ($K_m/V_{max}$) = 0.1740 |
| 0.08 | 0.136 | 7.35 | 12.50 | y = 5.2138 |
| 0.06 | 0.123 | 8.13 | 16.67 | x = -29.9715 |
| 0.04 | 0.104 | 9.62 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.072 | 13.89 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.140 | 7.14 | 10.00 | Slope ($K_m/V_{max}$) = 0.1944 |
| 0.08 | 0.131 | 7.63 | 12.50 | y = 5.1600 |
| 0.06 | 0.119 | 8.40 | 16.67 | x = -26.5492 |
| 0.04 | 0.101 | 9.90 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.067 | 14.92 | 50.00 | |

Table III.17 Lineweaver-Burk data of compound 16 (trypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
| 0.10 | 0.132 | 7.57 | 10.00 | Slope ($K_m/V_{max}$) = 0.3421 |
| 0.08 | 0.116 | 8.62 | 12.50 | y = 4.1378 |
| 0.06 | 0.104 | 9.62 | 16.67 | x = -12.0941 |
| 0.04 | 0.079 | 12.66 | 25.00 | Correlation coefficient = 0.9996 |
| 0.02 | 0.047 | 21.28 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.104 | 9.62 | 10.00 | Slope ($K_m/V_{max}$) = 0.3458 |
| 0.08 | 0.098 | 10.20 | 12.50 | y = 6.0735 |
| 0.06 | 0.086 | 11.63 | 16.67 | x = -17.5625 |
| 0.04 | 0.066 | 15.15 | 25.00 | Correlation coefficient = 0.9989 |
| 0.02 | 0.043 | 23.25 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.083 | 12.05 | 10.00 | Slope ($K_m/V_{max}$) = 0.4345 |
| 0.08 | 0.076 | 13.16 | 12.50 | y = 7.7598 |
| 0.06 | 0.067 | 14.92 | 16.67 | x = -17.0000 |
| 0.04 | 0.053 | 18.87 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.034 | 29.41 | 50.00 | |

Table III.18 Lineweaver-Burk data of compound 17 (trypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
| 0.10 | 0.166 | 6.02 | 10.00 | Slope $(K_m/V_{max})$ = 0.2078 |
| 0.08 | 0.156 | 6.41 | 12.50 | y = 3.8692 |
| 0.06 | 0.136 | 7.35 | 16.67 | x = -18.6201 |
| 0.04 | 0.111 | 9.01 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.070 | 14.28 | 50.00 | |
| Inhibitor = 150 $\mu$l. | | | | |
| 0.10 | 0.112 | 8.93 | 10.00 | Slope $(K_m/V_{max})$ = 0.2670 |
| 0.08 | 0.104 | 9.62 | 12.50 | y = 6.3170 |
| 0.06 | 0.093 | 10.75 | 16.67 | x = -23.6518 |
| 0.04 | 0.076 | 13.16 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.051 | 19.61 | 50.00 | |
| Inhibitor = 200 $\mu$l. | | | | |
| 0.10 | 0.080 | 12.50 | 10.00 | Slope $(K_m/V_{max})$ = 0.3627 |
| 0.08 | 0.074 | 13.51 | 12.50 | y = 8.9451 |
| 0.06 | 0.067 | 14.92 | 16.67 | x = -24.6598 |
| 0.04 | 0.055 | 18.18 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.037 | 27.03 | 50.00 | |

Table III.19 Lineweaver-Burk data of compound 1 (chymotrypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| $BAN\times10^{-3}$ | V | 1/V | $1/BAN\times10^{3}$ | Regression Analysis |
| 0.10 | 0.176 | 5.68 | 10.00 | Slope ($K_m/V_{max}$) = 0.0950 |
| 0.08 | 0.168 | 5.95 | 12.50 | y = 4.7218 |
| 0.06 | 0.157 | 6.37 | 16.67 | x = -49.6800 |
| 0.04 | 0.144 | 6.94 | 25.00 | Correlation coefficient = 0.9983 |
| 0.02 | 0.105 | 9.52 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.149 | 6.71 | 10.00 | Slope ($K_m/V_{max}$) = 0.0984 |
| 0.08 | 0.144 | 6.94 | 12.50 | y = 5.7075 |
| 0.06 | 0.136 | 7.35 | 16.67 | x = -58.0136 |
| 0.04 | 0.123 | 8.13 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.094 | 10.64 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.106 | 9.43 | 10.00 | Slope ($K_m/V_{max}$) = 0.1014 |
| 0.08 | 0.104 | 9.62 | 12.50 | y = 8.3619 |
| 0.06 | 0.098 | 10.20 | 16.67 | x = -82.4366 |
| 0.04 | 0.094 | 10.63 | 25.00 | Correlation coefficient = 0.9953 |
| 0.02 | 0.074 | 13.51 | 50.00 | |

Table III.20 Lineweaver-Burk data of compound 2 (chymotrypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
| 0.10 | 0.130 | 7.69 | 10.00 | Slope ($K_m/V_{max}$) = 0.1748 |
| 0.08 | 0.123 | 8.13 | 12.50 | y = 5.9607 |
| 0.06 | 0.112 | 8.93 | 16.67 | x = -34.1006 |
| 0.04 | 0.097 | 10.31 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.067 | 14.70 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.117 | 8.54 | 10.00 | Slope ($K_m/V_{max}$) = 0.1764 |
| 0.08 | 0.111 | 9.01 | 12.50 | y = 6.8151 |
| 0.06 | 0.102 | 9.80 | 16.67 | x = -38.6435 |
| 0.04 | 0.089 | 11.24 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.064 | 15.62 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.101 | 9.90 | 10.00 | Slope ($K_m/V_{max}$) = 0.1694 |
| 0.08 | 0.097 | 10.31 | 12.50 | y = 8.1738 |
| 0.06 | 0.091 | 10.99 | 16.67 | x = -48.2507 |
| 0.04 | 0.081 | 12.34 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.060 | 16.67 | 50.00 | |

Table III.21 Lineweaver-Burk data of compound 3 (chymotrypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| BANx$10^{-3}$ | V | 1/V | 1/BANx$10^{3}$ | Regression Analysis |
| 0.10 | 0.120 | 8.33 | 10.00 | Slope ($K_m/V_{max}$) = 0.4040 |
| 0.08 | 0.116 | 8.62 | 12.50 | y = 3.4424 |
| 0.06 | 0.102 | 9.80 | 16.67 | x = -8.5203 |
| 0.04 | 0.080 | 12.50 | 25.00 | Correlation coefficient = 0.9938 |
| 0.02 | 0.0415 | 24.09 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.069 | 14.49 | 10.00 | Slope ($K_m/V_{max}$) = 0.5719 |
| 0.08 | 0.063 | 15.07 | 12.50 | y = 8.4441 |
| 0.06 | 0.055 | 18.18 | 16.67 | x = -14.7666 |
| 0.04 | 0.044 | 22.73 | 25.00 | Correlation coefficient = 0.9994 |
| 0.02 | 0.027 | 37.04 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.051 | 19.61 | 10.00 | Slope ($K_m/V_{max}$) = 0.6516 |
| 0.08 | 0.047 | 21.28 | 12.50 | y = 13.0747 |
| 0.06 | 0.042 | 23.81 | 16.67 | x = -20.0665 |
| 0.04 | 0.034 | 29.41 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.021 | 47.62 | 50.00 | |

Table III.22 Lineweaver-Burk data of compound 4 (chymotrypsin)

| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
|---|---|---|---|---|
| Inhibitor = 0 | | | | |
| 0.10 | 0.152 | 6.58 | 10.00 | Slope $(K_m/V_{max})$ = 0.1069 |
| 0.08 | 0.146 | 6.85 | 12.50 | y = 5.5283 |
| 0.06 | 0.136 | 7.35 | 16.67 | x = -51.6975 |
| 0.04 | 0.122 | 8.20 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.092 | 10.87 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.136 | 7.35 | 10.00 | Slope $(K_m/V_{max})$ = 0.1111 |
| 0.08 | 0.131 | 7.63 | 12.50 | y = 6.2018 |
| 0.06 | 0.126 | 7.94 | 16.67 | x = -55.8360 |
| 0.04 | 0.111 | 9.01 | 25.00 | Correlation coefficient = 0.9994 |
| 0.02 | 0.085 | 11.76 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.121 | 8.26 | 10.00 | Slope $(K_m/V_{max})$ = 0.1222 |
| 0.08 | 0.115 | 8.69 | 12.50 | y = 7.0388 |
| 0.06 | 0.112 | 8.93 | 16.67 | x = -57.6236 |
| 0.04 | 0.099 | 10.10 | 25.00 | Correlation coefficient = 0.9988 |
| 0.02 | 0.076 | 13.16 | 50.00 | |

Table III.23 Lineweaver-Burk data of compound 5 (chymotrypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| $BAN\times10^{-3}$ | V | 1/V | $1/BAN\times10^{3}$ | Regression Analysis |
| 0.10 | 0.143 | 6.99 | 10.00 | Slope ($K_m/V_{max}$) = 0.1014 |
| 0.08 | 0.138 | 7.25 | 12.50 | y = 6.0036 |
| 0.06 | 0.133 | 7.52 | 16.67 | x = -59.1805 |
| 0.04 | 0.113 | 8.85 | 25.00 | Correlation coefficient = 0.9938 |
| 0.02 | 0.091 | 10.99 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.116 | 8.62 | 10.00 | Slope ($K_m/V_{max}$) = 0.1196 |
| 0.08 | 0.114 | 8.77 | 12.50 | y = 7.3646 |
| 0.06 | 0.107 | 9.34 | 16.67 | x = -61.5662 |
| 0.04 | 0.096 | 10.42 | 25.00 | Correlation coefficient = 0.9995 |
| 0.02 | 0.075 | 13.33 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.092 | 10.87 | 10.00 | Slope ($K_m/V_{max}$) = 0.1448 |
| 0.08 | 0.089 | 11.24 | 12.50 | y = 9.4618 |
| 0.06 | 0.084 | 11.90 | 16.67 | x = -65.3459 |
| 0.04 | 0.076 | 13.16 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.060 | 16.67 | 50.00 | |

Table III.24 Lineweaver-Burk data of compound 6 (chymotrypsin)

| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
|---|---|---|---|---|
| Inhibitor = 0 | | | | |
| 0.10 | 0.139 | 7.19 | 10.00 | Slope ($K_m/V_{max}$) = 0.1461 |
| 0.08 | 0.134 | 7.46 | 12.50 | y = 5.6385 |
| 0.06 | 0.124 | 8.06 | 16.67 | x = -38.5997 |
| 0.04 | 0.109 | 9.17 | 25.00 | Correlation coefficient = 0.9994 |
| 0.02 | 0.077 | 12.99 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.158 | 8.40 | 10.00 | Slope ($K_m/V_{max}$) = 0.1585 |
| 0.08 | 0.119 | 8.77 | 12.50 | y = 6.7587 |
| 0.06 | 0.114 | 9.43 | 16.67 | x = -42.6304 |
| 0.04 | 0.094 | 10.63 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.068 | 14.71 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.099 | 10.10 | 10.00 | Slope ($K_m/V_{max}$) = 0.1750 |
| 0.08 | 0.096 | 10.42 | 12.50 | y = 8.3290 |
| 0.06 | 0.089 | 11.24 | 16.67 | x = -47.6049 |
| 0.04 | 0.078 | 12.82 | 25.00 | Correlation coefficient = 0.9996 |
| 0.02 | 0.057 | 17.04 | 50.00 | |

Table III.25 Lineweaver-Burk data of compound 7 (chymotrypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| BANx$10^{-3}$ | V | 1/V | 1/BANx$10^{3}$ | Regression Analysis |
| 0.10 | 0.104 | 9.62 | 10.00 | Slope ($K_m/V_{max}$) = 0.1237 |
| 0.08 | 0.102 | 9.80 | 12.50 | y = 8.3937 |
| 0.06 | 0.096 | 10.42 | 16.67 | x = -67.8618 |
| 0.04 | 0.085 | 11.76 | 25.00 | Correlation coefficient = 0.9968 |
| 0.02 | 0.069 | 14.49 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.091 | 10.99 | 10.00 | Slope ($K_m/V_{max}$) = 0.1413 |
| 0.08 | 0.088 | 11.36 | 12.50 | y = 9.6202 |
| 0.06 | 0.083 | 12.05 | 16.67 | x = -68.0958 |
| 0.04 | 0.076 | 13.16 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.060 | 16.67 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.076 | 13.16 | 10.00 | Slope ($K_m/V_{max}$) = 0.1656 |
| 0.08 | 0.075 | 13.33 | 12.50 | y = 11.4810 |
| 0.06 | 0.071 | 14.08 | 16.67 | x = -69.3358 |
| 0.04 | 0.062 | 16.13 | 25.00 | Correlation coefficient = 0.9939 |
| 0.02 | 0.051 | 19.61 | 50.00 | |

Table III.26 Lineweaver-Burk data of compound 8 (chymotrypsin)

| $BAN \times 10^{-3}$ | V | 1/V | $1/BAN \times 10^{3}$ | Regression Analysis |
|---|---|---|---|---|
| Inhibitor = 0 | | | | |
| 0.10 | 0.197 | 5.08 | 10.00 | Slope ($K_m/V_{max}$) = 0.0869 |
| 0.08 | 0.189 | 5.29 | 12.50 | y = 4.2111 |
| 0.06 | 0.177 | 5.65 | 16.67 | x = -48.4440 |
| 0.04 | 0.156 | 6.41 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.117 | 8.55 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.150 | 6.67 | 10.00 | Slope ($K_m/V_{max}$) = 0.1016 |
| 0.08 | 0.144 | 6.94 | 12.50 | y = 5.6855 |
| 0.06 | 0.135 | 7.41 | 16.67 | x = -55.9445 |
| 0.04 | 0.121 | 8.26 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.093 | 10.75 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.132 | 7.58 | 10.00 | Slope ($K_m/V_{max}$) = 0.1087 |
| 0.08 | 0.128 | 7.81 | 12.50 | y = 6.4774 |
| 0.06 | 0.121 | 8.26 | 16.67 | x = -59.5703 |
| 0.04 | 0.108 | 9.26 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.084 | 11.90 | 50.00 | |

Table III.27 Lineweaver-Burk data of compound 9 (chymotrypsin)

| | | | | |
|---|---|---|---|---|
| Inhibitor = 0 | | | | |
| BANx$10^{-3}$ | V | 1/V | 1/BANx$10^{3}$ | Regression Analysis |
| 0.10 | 0.132 | 7.58 | 10.00 | Slope ($K_m/V_{max}$) = 0.1233 |
| 0.08 | 0.124 | 8.06 | 12.50 | y = 6.4850 |
| 0.06 | 0.114 | 8.77 | 16.67 | x = -52.6029 |
| 0.04 | 0.106 | 9.43 | 25.00 | Correlation coefficient = 0.9971 |
| 0.02 | 0.079 | 12.66 | 50.00 | |
| Inhibitor = 150 μl. | | | | |
| 0.10 | 0.121 | 8.26 | 10.00 | Slope ($K_m/V_{max}$) = 0.1673 |
| 0.08 | 0.116 | 8.62 | 12.50 | y = 6.4787 |
| 0.06 | 0.108 | 9.26 | 16.67 | x = -38.7335 |
| 0.04 | 0.096 | 10.42 | 25.00 | Correlation coefficient = 0.9986 |
| 0.02 | 0.067 | 14.93 | 50.00 | |
| Inhibitor = 200 μl. | | | | |
| 0.10 | 0.113 | 8.85 | 10.00 | Slope ($K_m/V_{max}$) = 0.1888 |
| 0.08 | 0.106 | 9.43 | 12.50 | y = 6.9308 |
| 0.06 | 0.101 | 9.90 | 16.67 | x = -36.7084 |
| 0.04 | 0.086 | 11.63 | 25.00 | Correlation coefficient = 0.9993 |
| 0.02 | 0.061 | 16.40 | 50.00 | |

Table III.28 Lineweaver-Burk data of compound 10 (chymotrypsin)

| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
|---|---|---|---|---|
| **Inhibitor = 0** | | | | |
| 0.10 | 0.127 | 7.87 | 10.00 | Slope ($K_m/V_{max}$) = 0.1611 |
| 0.08 | 0.122 | 8.20 | 12.50 | y = 6.2401 |
| 0.06 | 0.112 | 8.93 | 16.67 | x = -38.7416 |
| 0.04 | 0.097 | 10.31 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.070 | 14.28 | 50.00 | |
| **Inhibitor = 150 µl.** | | | | |
| 0.10 | 0.100 | 10.00 | 10.00 | Slope ($K_m/V_{max}$) = 0.1926 |
| 0.08 | 0.093 | 10.75 | 12.50 | y = 8.1851 |
| 0.06 | 0.087 | 11.49 | 16.67 | x = -42.5994 |
| 0.04 | 0.078 | 12.82 | 25.00 | Correlation coefficient = 0.9990 |
| 0.02 | 0.056 | 17.86 | 50.00 | |
| **Inhibitor = 200 µl.** | | | | |
| 0.10 | 0.084 | 11.90 | 10.00 | Slope ($K_m/V_{max}$) = 0.2250 |
| 0.08 | 0.082 | 12.19 | 12.50 | y = 9.5790 |
| 0.06 | 0.074 | 13.51 | 16.67 | x = -42.5787 |
| 0.04 | 0.066 | 15.15 | 25.00 | Correlation coefficient = 0.9992 |
| 0.02 | 0.048 | 20.83 | 50.00 | |

Table III.29 Lineweaver-Burk data of compound 11 (chymotrypsin)

| $BAN\times10^{-3}$ | V | 1/V | $1/BAN\times10^{3}$ | Regression Analysis |
|---|---|---|---|---|
| Inhibitor = 0 | | | | |
| 0.10 | 0.149 | 6.71 | 10.00 | Slope ($K_m/V_{max}$) = 0.0799 |
| 0.08 | 0.146 | 6.85 | 12.50 | y = 5.9011 |
| 0.06 | 0.137 | 7.30 | 16.67 | x = -73.8397 |
| 0.04 | 0.127 | 7.87 | 25.00 | Correlation coefficient = 0.9994 |
| 0.02 | 0.101 | 9.90 | 50.00 | |
| Inhibitor = 150 μl. | | | | |
| 0.10 | 0.145 | 6.91 | 10.00 | Slope ($K_m/V_{max}$) = 0.0997 |
| 0.08 | 0.141 | 7.09 | 12.50 | y = 5.8765 |
| 0.06 | 0.132 | 7.57 | 16.67 | x = -58.9698 |
| 0.04 | 0.120 | 8.33 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.092 | 10.87 | 50.00 | |
| Inhibitor = 200 μl. | | | | |
| 0.10 | 0.136 | 7.35 | 10.00 | Slope ($K_m/V_{max}$) = 0.1490 |
| 0.08 | 0.130 | 7.69 | 12.50 | y = 5.8706 |
| 0.06 | 0.118 | 8.47 | 16.67 | x = -39.4098 |
| 0.04 | 0.105 | 9.52 | 25.00 | Correlation coefficient = 0.9995 |
| 0.02 | 0.075 | 13.33 | 50.00 | |

Table III.30 Lineweaver-Burk data of compound 12 (chymotrypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| BANx$10^{-3}$ | V | 1/V | 1/BANx$10^{3}$ | Regression Analysis |
| 0.10 | 0.126 | 7.94 | 10.00 | Slope ($K_m/V_{max}$) = 0.1137 |
| 0.08 | 0.121 | 8.26 | 12.50 | y = 6.8068 |
| 0.06 | 0.115 | 8.69 | 16.67 | x = -59.8898 |
| 0.04 | 0.104 | 9.62 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.080 | 12.50 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.120 | 8.33 | 10.00 | Slope ($K_m/V_{max}$) = 0.1550 |
| 0.08 | 0.116 | 8.62 | 12.50 | y = 6.7424 |
| 0.06 | 0.107 | 9.34 | 16.67 | x = -43.4959 |
| 0.04 | 0.094 | 10.63 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.069 | 14.49 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.116 | 8.62 | 10.00 | Slope ($K_m/V_{max}$) = 0.1761 |
| 0.08 | 0.111 | 9.01 | 12.50 | y = 6.8179 |
| 0.06 | 0.103 | 9.71 | 16.67 | x = -38.7057 |
| 0.04 | 0.089 | 11.23 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.064 | 15.63 | 50.00 | |

Table III.31 Lineweaver-Burk data of compound 13 (chymotrypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
| 0.10 | 0.147 | 6.80 | 10.00 | Slope ($K_m/V_{max}$) = 0.1380 |
| 0.08 | 0.139 | 7.19 | 12.50 | y = 5.4503 |
| 0.06 | 0.129 | 7.75 | 16.67 | x = -39.4881 |
| 0.04 | 0.112 | 8.93 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.081 | 12.34 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.138 | 7.25 | 10.00 | Slope ($K_m/V_{max}$) = 0.1872 |
| 0.08 | 0.131 | 7.63 | 12.50 | y = 5.3178 |
| 0.06 | 0.118 | 8.47 | 16.67 | x = -28.4098 |
| 0.04 | 0.101 | 9.90 | 25.00 | Correlation coefficient = 0.9998 |
| 0.02 | 0.068 | 14.71 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.131 | 7.63 | 10.00 | Slope ($K_m/V_{max}$) = 0.2401 |
| 0.08 | 0.122 | 8.20 | 12.50 | y = 5.2048 |
| 0.06 | 0.108 | 9.26 | 16.67 | x = -21.6743 |
| 0.04 | 0.090 | 11.11 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.058 | 17.24 | 50.00 | |

Table III.32 Lineweaver-Burk data of compound 14 (chymotrypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
| 0.10 | 0.146 | 6.85 | 10.00 | Slope ($K_m/V_{max}$) = 0.1430 |
| 0.08 | 0.138 | 7.25 | 12.50 | y = 5.5141 |
| 0.06 | 0.123 | 8.13 | 16.67 | x = -38.5533 |
| 0.04 | 0.111 | 9.01 | 25.00 | Correlation coefficient = 0.9983 |
| 0.02 | 0.079 | 12.66 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.135 | 7.41 | 10.00 | Slope ($K_m/V_{max}$) = 0.1983 |
| 0.08 | 0.126 | 7.94 | 12.50 | y = 5.4713 |
| 0.06 | 0.113 | 8.85 | 16.67 | x = -27.5869 |
| 0.04 | 0.096 | 10.42 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.065 | 15.38 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.123 | 8.13 | 10.00 | Slope ($K_m/V_{max}$) = 0.2671 |
| 0.08 | 0.113 | 8.85 | 12.50 | y = 5.5320 |
| 0.06 | 0.099 | 10.10 | 16.67 | x = -20.7144 |
| 0.04 | 0.082 | 12.20 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.053 | 18.87 | 50.00 | |

Table III.33 Lineweaver-Burk data of compound 15 (chymotrypsin)

| Inhibitor = 0 | | | | |
|---|---|---|---|---|
| $BAN \times 10^{-3}$ | V | 1/V | $1/BAN \times 10^{3}$ | Regression Analysis |
| 0.10 | 0.159 | 6.29 | 10.00 | Slope ($K_m/V_{max}$) = 0.1304 |
| 0.08 | 0.151 | 6.62 | 12.50 | y = 5.0109 |
| 0.06 | 0.140 | 7.14 | 16.67 | x = -38.4329 |
| 0.04 | 0.119 | 8.40 | 25.00 | Correlation coefficient = 0.9994 |
| 0.02 | 0.087 | 11.49 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.147 | 6.80 | 10.00 | Slope ($K_m/V_{max}$) = 0.1612 |
| 0.08 | 0.142 | 7.04 | 12.50 | y = 5.0595 |
| 0.06 | 0.130 | 7.69 | 16.67 | x = -31.3892 |
| 0.04 | 0.111 | 9.01 | 25.00 | Correlation coefficient = 0.9995 |
| 0.02 | 0.076 | 13.16 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.132 | 7.57 | 10.00 | Slope ($K_m/V_{max}$) = 0.2360 |
| 0.08 | 0.124 | 8.06 | 12.50 | y = 5.1278 |
| 0.06 | 0.111 | 9.01 | 16.67 | x = -21.7307 |
| 0.04 | 0.091 | 10.99 | 25.00 | Correlation coefficient = 0.9999 |
| 0.02 | 0.059 | 16.95 | 50.00 | |

Table III.34 Lineweaver-Burk data of compound 16 (chymotrypsin)

| $BANx10^{-3}$ | V | 1/V | $1/BANx10^{3}$ | Regression Analysis |
|---|---|---|---|---|
| Inhibitor = 0 | | | | |
| 0.10 | 0.183 | 5.46 | 10.00 | Slope ($K_m/V_{max}$) = 0.1491 |
| 0.08 | 0.161 | 6.21 | 12.50 | y = 4.2364 |
| 0.06 | 0.146 | 6.85 | 16.67 | x = -28.4048 |
| 0.04 | 0.124 | 8.06 | 25.00 | Correlation coefficient = 0.9976 |
| 0.02 | 0.086 | 11.63 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.117 | 8.55 | 10.00 | Slope ($K_m/V_{max}$) = 0.1837 |
| 0.08 | 0.113 | 8.85 | 12.50 | y = 6.6864 |
| 0.06 | 0.101 | 9.90 | 16.67 | x = -36.4075 |
| 0.04 | 0.089 | 11.23 | 25.00 | Correlation coefficient = o.9994 |
| 0.02 | 0.063 | 15.87 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.081 | 12.34 | 10.00 | Slope ($K_m/V_{max}$) = 0.2394 |
| 0.08 | 0.079 | 12.66 | 12.50 | y = 9.8478 |
| 0.06 | 0.073 | 13.70 | 16.67 | x = -41.1374 |
| 0.04 | 0.062 | 16.13 | 25.00 | Correlation coefficient = 0.9987 |
| 0.02 | 0.046 | 21.74 | 50.00 | |

Table III.35 Lineweaver-Burk data of compound 17 (chymotrypsin)

| BANx$10^{-3}$ | V | 1/V | 1/BANx$10^{3}$ | Regression Analysis |
|---|---|---|---|---|
| Inhibitor = 0 | | | | |
| 0.10 | 0.151 | 6.62 | 10.00 | Slope ($K_m/V_{max}$) = 0.1398 |
| 0.08 | 0.138 | 7.25 | 12.50 | y = 5.4464 |
| 0.06 | 0.128 | 7.81 | 16.67 | x = -38.9659 |
| 0.04 | 0.109 | 9.17 | 25.00 | Correlation coefficient = 0.9972 |
| 0.02 | 0.081 | 12.34 | 50.00 | |
| Inhibitor = 150 µl. | | | | |
| 0.10 | 0.102 | 9.80 | 10.00 | Slope ($K_m/V_{max}$) = 0.1796 |
| 0.08 | 0.098 | 10.20 | 12.50 | y = 8.0192 |
| 0.06 | 0.091 | 10.99 | 16.67 | x = -44.6524 |
| 0.04 | 0.079 | 12.66 | 25.00 | Correlation coefficient = 0.9996 |
| 0.02 | 0.059 | 16.95 | 50.00 | |
| Inhibitor = 200 µl. | | | | |
| 0.10 | 0.084 | 11.90 | 10.00 | Slope ($K_m/V_{max}$) = 0.2066 |
| 0.08 | 0.079 | 12.66 | 12.50 | y = 10.1674 |
| 0.06 | 0.071 | 14.08 | 16.67 | x = -49.2022 |
| 0.04 | 0.065 | 15.38 | 25.00 | Correlation coefficient = 0.9961 |
| 0.02 | 0.049 | 20.41 | 50.00 | |

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