Enzyme Kinetics Analysis Report

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1 Introduction

Enzyme kinetics is the study of the chemical reactions that are catalyzed by enzymes. This report analyzes two sets of experimental data on enzyme kinetics, one with an inhibitor and one without an inhibitor. Calculates the V_{max}, K_M and K_I values.

2 Input data

(a) Reaction without inhibitor

[S](mmol/l)	[I](mmol/l)	V (µmol/L/min)
1	0	0.67
2	0	1.13
3	0	1.48
4	0	1.74
5	0	1.98

(b) Reaction with inhibitor

[S](mmol/l)	[I](mmol/l)	V (µmol/L/min)
1	1	0.05
2	1	0.10
3	1	0.15
4	1	0.16
5	1	0.18

Table 1: Reaction rates with and without inhibitor

Equations

The Michaelis-Menten equation is given by:

$$V = V_{max} \frac{[S]}{K_M + [S]} \tag{1}$$

The Lineweaver-Burk equation is given by:

$$\frac{1}{V} = \frac{1}{V_{max}} + \frac{1}{K_M} \cdot \frac{1}{[S]} \tag{2}$$

The Michaelis-Menten equation can be derived from the Lineweaver-Burk equation by multiplying both sides by V and rearranging the terms:

$$V = \frac{V_{max}}{1 + \frac{K_M}{[S]}} \tag{3}$$

The Michaelis-Menten equation can be used to calculate the K_I value, in noncompetitive inhibition, as follows:

$$V = \frac{\frac{V_{max}}{1 + \frac{[I]}{K_I}} \cdot [S]}{K_M + [S]} \tag{4}$$

Plots

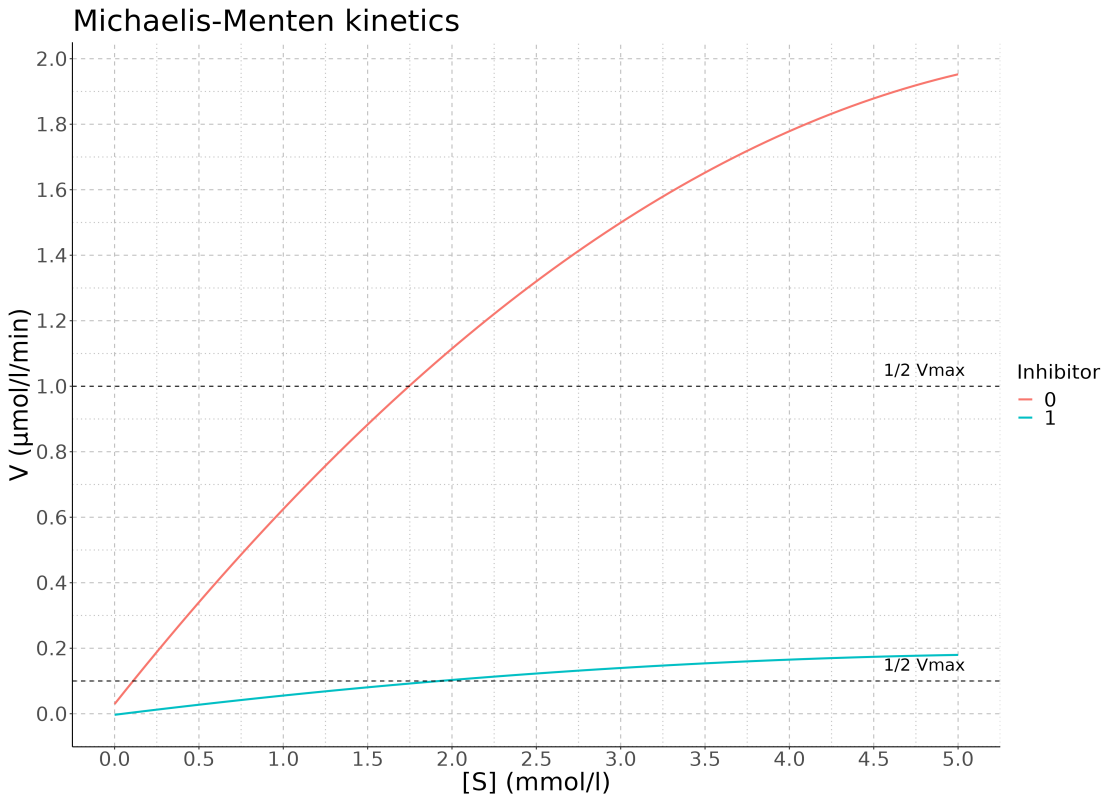

Figure 1: Michaelis-Menten kinetics plot showing the relationship between substrate concentration and reaction rate with and without inhibitor. The plot also includes fitted values and 1/2 V_{max} lines for both datasets.

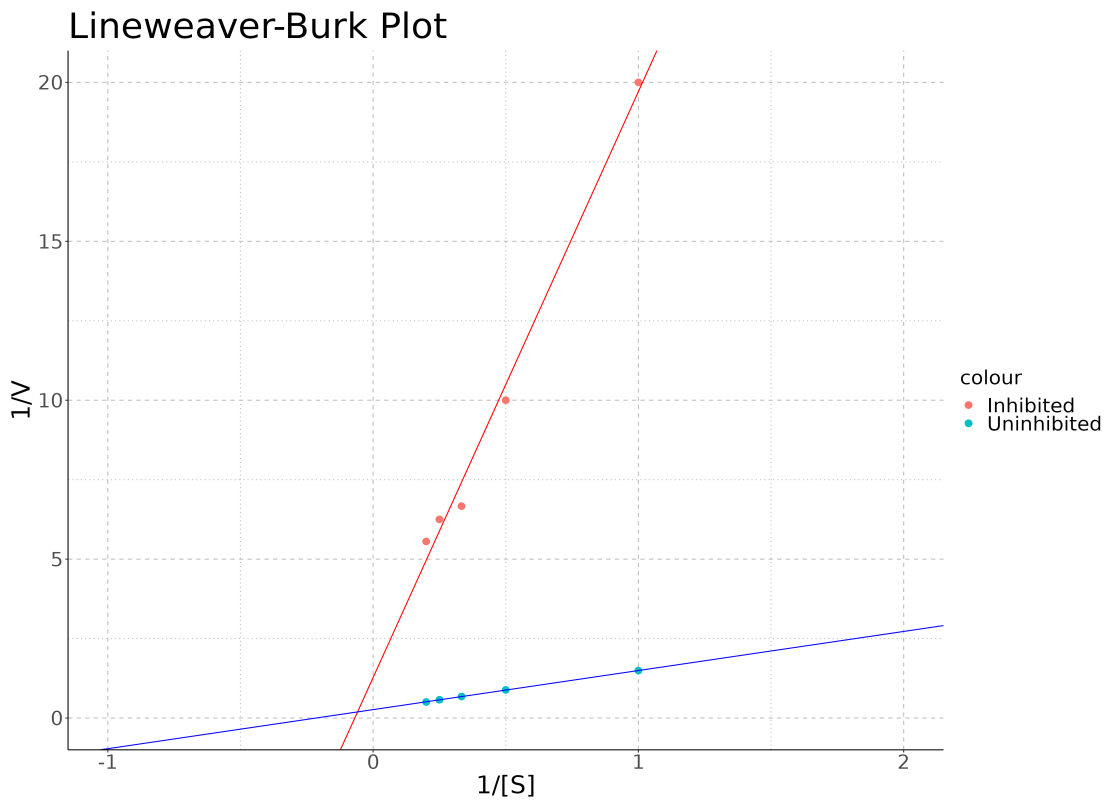

Figure 2: Lineweaver-Burk plot showing the reciprocal of substrate concentration ($1/[S]$) versus the reciprocal of enzyme velocity ($1/V$) for inhibited (red) and uninhibited (blue) enzyme reactions. The plot includes two linear regression lines with slopes and intercepts calculated from the data.

3 Uninhibited Reaction

The first set of data is from an uninhibited reaction. The data was fit to the Michaelis-Menten equation $V = V_{max}\frac{[S]}{K_M + [S]}$, where V_{max} is the maximum velocity of the reaction, $[S]$ is the substrate concentration, and K_M is the Michaelis constant. The fitted parameters are as follows:

- $V_{max} = 3.89 \pm 0.08$ mmol/l
- $K_M = 4.88 \pm 0.18$ µmol/L/min

Table 2: Model summary without inhibitor

	Estimate	Std. Error	t value	Pr($>|t|$)
V_{max}	3.892276	0.0846582	45.97636	0.0000227
K_M	4.881717	0.1828702	26.69498	0.0001153

Table 3: Confidence interval for Vm and Km for uninhibited reaction

	2.5%	97.5%
V_{max}	3.640811	4.185819
K_M	4.340821	5.518560

4 Inhibited Reaction

The second set of data is from an inhibited reaction. The data was fit to the Michaelis-Menten equation $V = V_{max}\frac{[S]}{K_M + [S]}$, where V_{max} is the maximum velocity of the reaction, $[S]$ is the substrate concentration, and K_M is the Michaelis constant. The fitted parameters are as follows:

- $V_{max} = 0.39 \pm 0.10$ mmol/l
- $K_M = 5.71 \pm 2.30$ µmol/L/min

Table 4: Model summary with inhibitor

	Estimate	Std. Error	t value	Pr($>\|t\|$)
V_{max}	0.3945488	0.0974732	4.047765	0.0271504
K_M	5.7125880	2.2981536	2.485729	0.0888187

Table 5: Confidence interval for V_{max} and K_M for inhibited reaction

	2.5%	97.5%
V_{max}	0.2437008	3.270304
K_M	2.5583919	74.920253

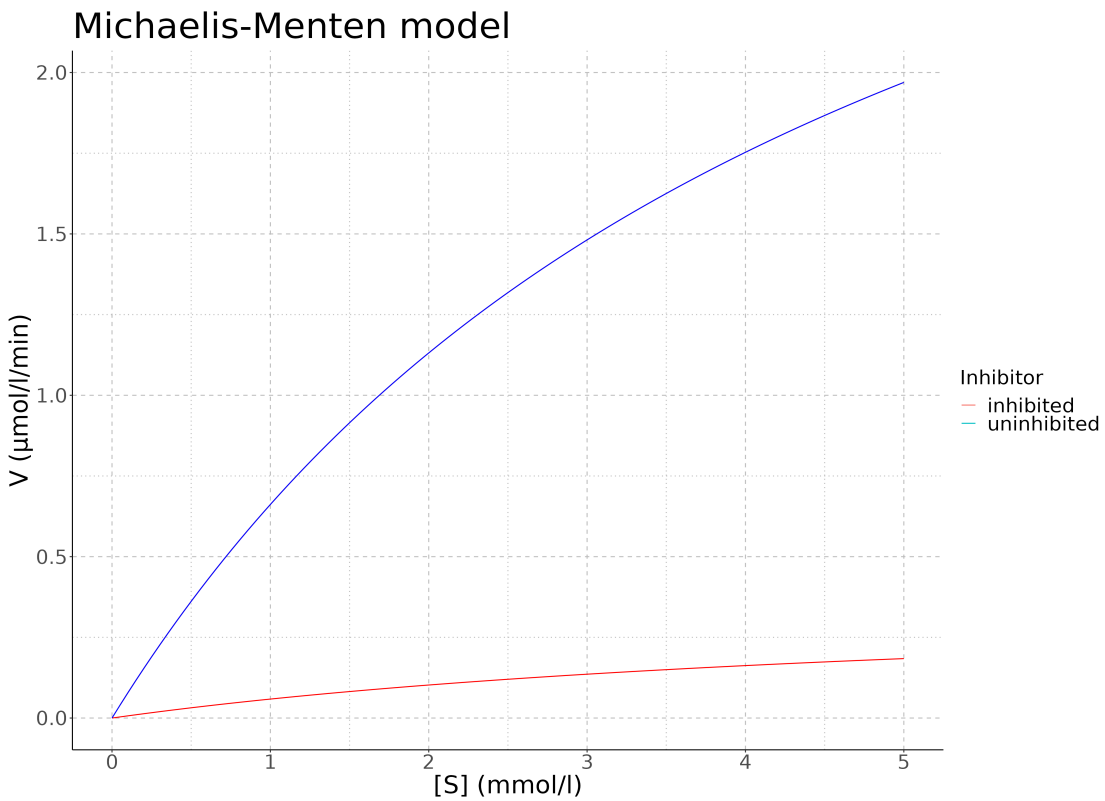

Figure 3: Michaelis-Menten model for enzyme kinetics, showing the substrate concentration [S] (mmol/l) on the x-axis and the reaction rate V (µmol/l/min) on the y-axis. The blue line represents the reaction rate without inhibitor, while the red line shows the reaction rate with inhibitor. The legend indicates whether the inhibitor is present or absent. The plot illustrates the effect of an inhibitor on the enzyme activity, with the inhibited reaction rate curve shifted downwards compared to the uninhibited reaction rate curve.

5 Non-competitive inhibition

The data was fit to the same Michaelis-Menten equation as before, but with an additional term to account for the inhibitor: $V = \dfrac{\dfrac{V_{max}}{1 + \frac{[I]}{K_I}} \cdot [S]}{K_M + [S]}$

where $[I]$ is the inhibitor concentration and K_I is the inhibition constant. The fitted parameters are as follows:

- $V_{max} = 0.39 \pm 0.10$ µmol/L/min
- $K_M = 5.71 \pm 2.30$ mmol/l
- $K_I = 0.10 \pm 0.00$ mmol/l

Table 6: Model summary for non-competitive inhibition

	Estimate	Std. Error	t value	Pr(> |t|)
V_{max}	3.8955119	0.0767059	50.78505	0
K_M	4.8887879	0.1656901	29.50562	0
K_I	0.1017605	0.0036314	28.02254	0

Table 7: Confidence interval for non-competitive inhibition

	2.5%	97.5%
V_{max}	3.7222470	4.0877450
K_M	4.5155594	5.3052748
K_I	0.0932454	0.1104207

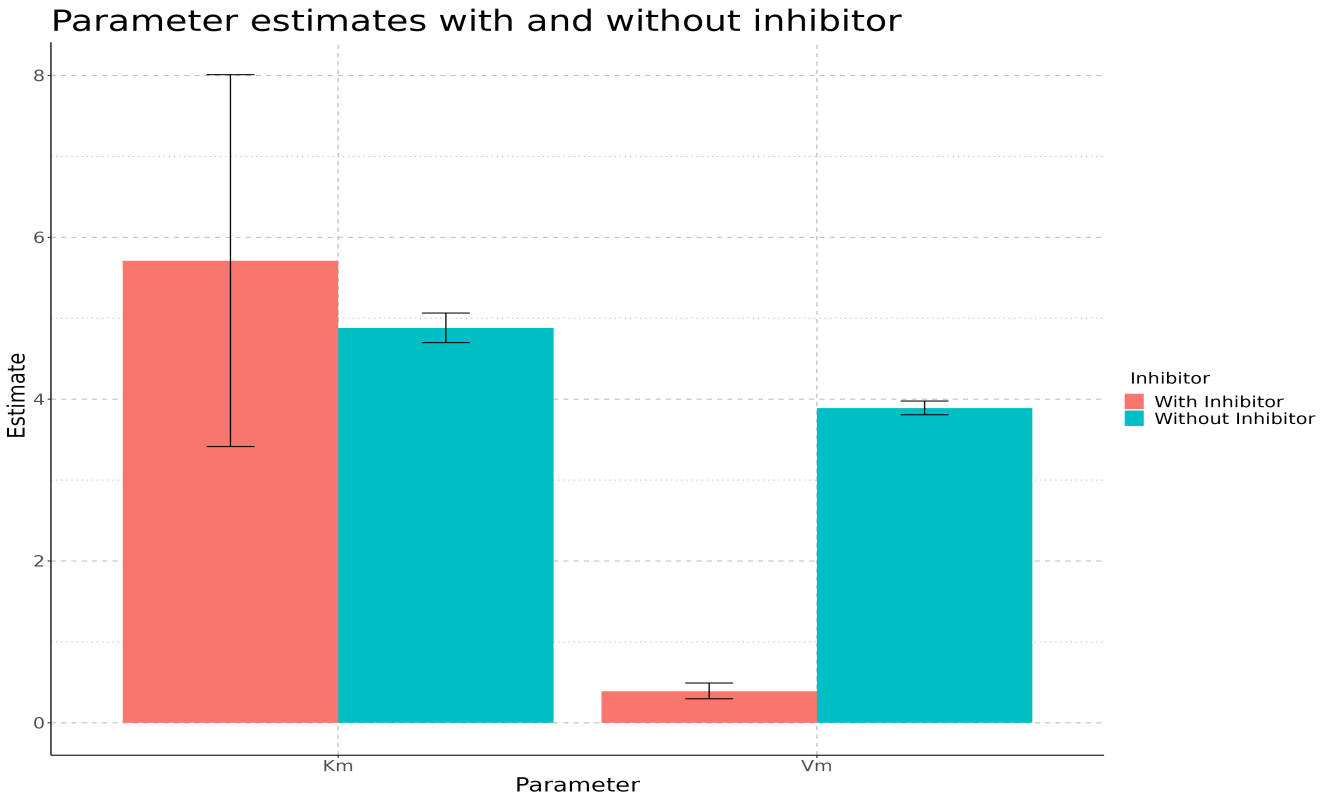

Figure 4: Bar plot showing parameter estimates with and without inhibitor for the V_{max} and K_M parameters. Error bars represent the standard error of the estimates.

6 Conclusion

As a result of enzyme kinetics analysis, it was found that the presence of inhibitor significantly reduced the maximum reaction rate (V_{max}) and slightly increased the Michaelis constant (K_M) compared to the reaction without inhibitor. From Lineweaver-Burk plot, it was determined that the inhibitor is non-competitive.

From Lineweaver-Burk plot, it was found that the inhibitor is non-competitive.

Inhibition of the reaction showed that the inhibitor could bind to the enzyme and prevent binding of the substrate, leading to a decrease in the reaction rate. An increase in K_M indicates that the inhibitor can alter the affinity of the enzyme for its substrate.

7 Code

The code for this project can be found at https://github.com/dodes24/enzymology
Code was written in R and jupyter notebook was used to create the project.
Latex was used to create the report.