Introduction to Bioinformatics
Simulation of Michaelis-Menten Kinetics

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A library for Stochastic Simulation of Michaelis-Menten Kinetics has been implemented by the author in C++ and the source code of the library along with the instructions of usage and a sample test program have been made available at `https://github.com/tadarsh/BT6090/tree/master/EnzymeKineticsLibrary`. The library also contains functions to fit a straight line and several other helper functions.

Introduction

Enzyme Kinetics

Consider the enzyme catalytic reaction,

$$E + S \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} ES \xrightarrow{k_2} E + P$$
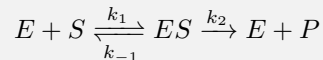
where E is the free enzyme, S is a substract, ES is the enzyme-substrate complex, and P is the product of the reaction.

Analytical Solution

Writing the rate equations,

$$\frac{d[ES]}{dt} = k_1[E][S] - k_{-1}[ES] - k_2[ES]$$

$$\frac{d[P]}{dt} = k_2[ES]$$

Initial Conditions

$$[E]_{t=0} = e_0$$
$$[S]_{t=0} = s_0$$
$$[ES]_{t=0} = 0$$
$$[P]_{t=0} = 0$$

Quasi Steady State Approximation

Assuming that the ES complex reaches a steady state much before the second reaction,

$$\frac{d[ES]}{dt} = 0$$

and $e_0 \ll s_0$, we get,

$$\frac{d[P]}{dt} = k_1[ES]_{steady} = \frac{k_2 e_0 s_0}{s_0 + \frac{k_{-1} + k_2}{k_1}}$$

rewriting the equation, we get,

$$\frac{d[P]}{dt} = k_1[ES]_{steady} = \frac{v_{max} s_0}{s_0 + k_m}$$

where,

$$v_{max} = k_2 e_0$$

is the maximum velocity of product formation and

$$k_m = \frac{k_{-1} + k_2}{k_1}$$

is defined as the Michaelis-Menten constant.

If we know the values of k_1, k_{-1}, k_2 and e_0, we can determine the constants k_m and $v_m ax$, using the quasi steady state approximation.

Aim of the simulation

The aim of the experiment is to determine the constants k_m and v_{max} by simulating the enzyme catalytic reaction without the quasi steady state approximation and comparing those values to values obtained using the quasi steady state approximation.
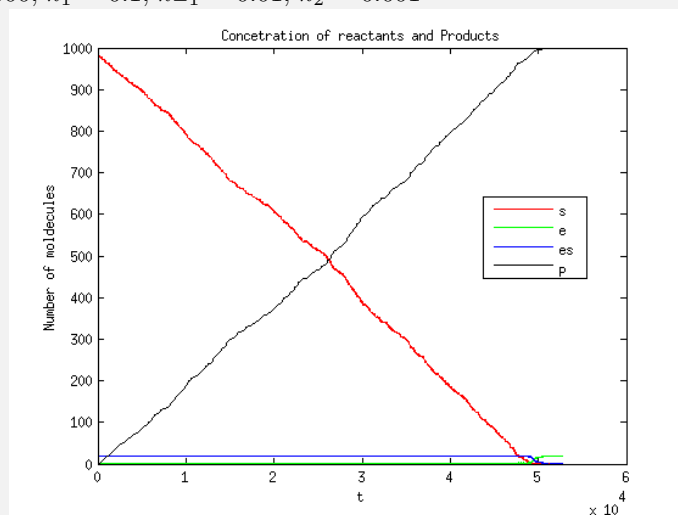
Reaction Simulation

The following procedure was adopted to simulate the reaction, assume initial values as stated above,

1. Generate two random numbers $r_1, r_2 \in (0, 1)$

2. Compute the reaction flux $rf = k_1(e)(s) + k_{-1}(es) + k_2(es)$

3. Find the time till next reaction $\tau = \frac{1}{rf} log \left(\frac{1}{r_1} \right)$

4. If $0 < r_2 \leq \frac{k_1(e)(s)}{rf}$, simulate reaction involving k_1 after time τ

5. If $\frac{k_1(e)(s)}{rf} < r_2 \leq \frac{k_1(e)(s)+k_{-1}(es)}{rf}$, simulate reaction involving k_{-1} after time τ

6. If $\frac{k_1(e)(s)+k_{-1}(es)}{rf} < r_2 < 1$, simulate reaction involving k_2 after time τ

7. Check for boundary conditions, $0 \leq e \leq e_0$,$0 \leq s \leq s_0$, $0 \leq es \leq e_0$, $0 \leq p \leq s_0$

8. Go to step 1, till $p < s_0$.

Concentration of reactants and products with the time

The simulation was performed and all the concentrations were plotting with respect to time.
For $e_0 = 20$, $s_0 = 1000$, $k_1 = 0.1$, $k_{-1} = 0.01$, $k_2 = 0.001$

Determining the Michaelis-Menten Constants

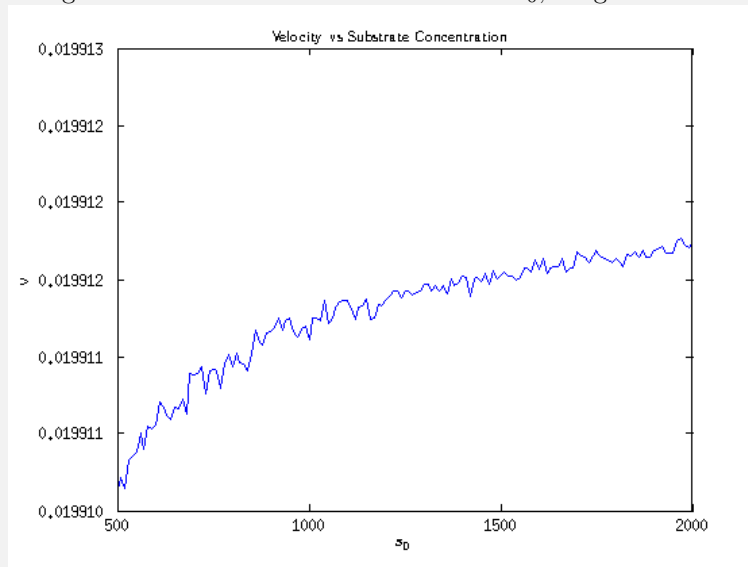The simulation was performed 100 times for each value of s_0 and several values of s_0, were used.

Calulation of Average Velocity

For a singe simulation, the average velocty

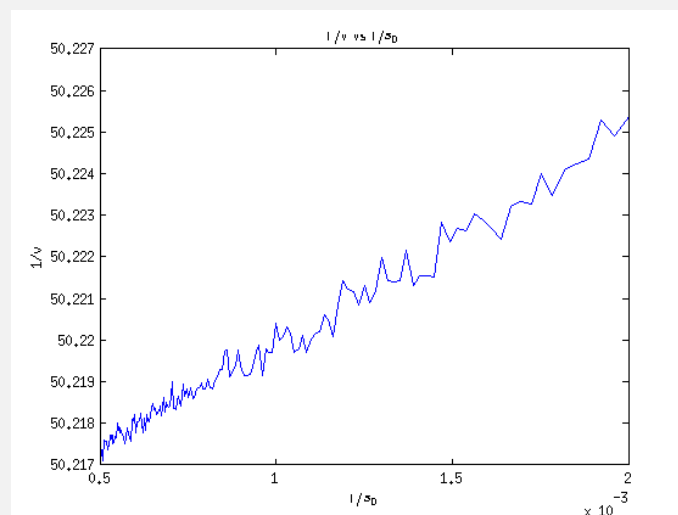$$v_{avg} = \frac{[P]_{final} - [P]_{initial}}{t_{final} - t_{initial}}$$

100 such values were calculated and averaged for each value of s_0.

On repeating the simulation for different values of s_0, we get the following plot.

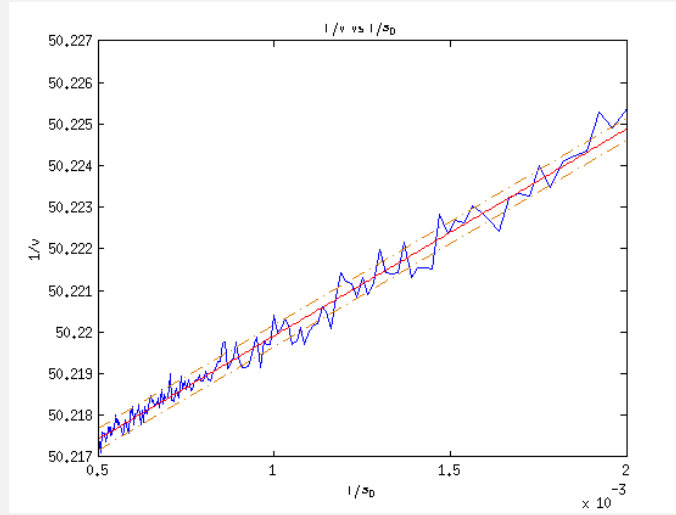Plotting the $\frac{1}{v}$ vs $\frac{1}{s_0}$, we get the following plot,

As mentioned previously, the velocity equation,

$$v = \frac{v_{max} s_0}{k_m + s_0}$$

rewriting the equation as,

$$\frac{1}{v} = \frac{1}{v_{max}} + \frac{k_m}{v_{max}}\left(\frac{1}{s_0}\right)$$

Fitting a straight line to the previous plot,

the equation which represents the straight line is the following,

$$\frac{1}{v} = 50.2149 + 4.9701\left(\frac{1}{s_0}\right)$$

Comparing the above two equations, we get

$$v_{max} = 0.0199 \pm 0.0004$$

$$k_m = 0.098 \pm 0.0082$$

Theoretically Computing Km and Vmax using QSSA

$$v_{max} = k_2 e_0 = 0.001 \times 20 = 0.02$$

$$k_m = \frac{k_{-1} + k_2}{k_1} = \frac{0.01 + 0.001}{0.1} = 0.11$$

Conclusion

The theoretical values of Michaelis-Menten constants were compared to the values obtained during simulation and were found to be similar.

The value of v_{max} obtained using simulation was $v_{max} = 0.0199$ compared to the theoretical value of 0.02. The value of $k_m = 0.098$ obtained using simulation is close to the theoretical value of 0.11.

References

[1] Wikipedia Article on Enzyme Kinetics

http://en.wikipedia.org/wiki/Enzyme_kinetics

[2] Tutorial on Michaelis-Menten Kinetics

http://www.wiley.com/college/pratt/0471393878/student/animations/enzyme_kinetics/
enzyme_kinetics.swf

[3] BT6090 Class Notes on Enzyme Kinetics