Give all answers to at least **4 significant figures**.

1. Characterizing the properties of enzymes is a very important endeavor in the biological sciences. Many enzymes follow Michaelis-Menten kinetics and have the following form for their reaction rates:

$$R([S]) = \frac{V_{max}[S]}{K_m + [S]},$$

where $[S]$ is the concentration of the substrate that the enzyme is catalyzing, $R([S])$ is the rate of production of the product, V_{max} is the maximum rate of the reaction (and depend on the enzyme concentration used, and K_m is the Michaelis-Menten constant, which characterizes a particular enzyme.

Below are data from Schmider, et. al. [1] for cytochrome P450 mediated demethylation of amitriptyline (AMI) to nortriptyline (N) by human liver microsomes.

[AMI] (μM)	N formation nmol/min/mg
5	0.20
10	0.45
15	0.60
25	0.80
50	1.35
100	2.17
200	2.68
260	2.89
330	3.05
500	3.12

From this table, we let $[S] = $ [AMI] and $R([S]) = $ N formation. We want to find the best values of V_{max} and K_m to characterize the enzyme cytochrome P450.

a. One of the easiest methods for finding the constants V_{max} and K_m is using the Lineweaver-Burk plot as discussed in lecture. To do this, you take the data and create a new table with the values $x = 1/[S]$ and $y = 1/R([S])$. Create this new table in Excel, then plot the data y vs x. Use Trendline to find the best fit to this straight line. The y-intercept has the value $1/V_{max}$ and the slope has the value K_m/V_{max}. Use this information to find V_{max} and K_m.

b. With this information, plot the original data with the Michaelis-Menten reaction formula for $[S] \in [0, 500]$. Find the prediction from the Michaelis-Menten reaction formula for the production of nortriptyline (N) when [AMI] is $[S] = 15, 50, 100, 200,$ and 500. Calculate the percent error between the model and the actual data. Find all intercepts and any asymptotes for this function for $[S] \geq 0$.

c. When a nonlinear least squares fit is applied to the data above, a better model is given

$$R([S]) = \frac{3.738[S]}{80.63 + [S]}.$$

With this information, plot the original data with this Michaelis-Menten reaction formula for $[S] \in [0, 500]$. Find the prediction from this Michaelis-Menten reaction formula for the production of nortriptyline (N) when [AMI] is $[S] = 15, 50, 100, 200$, and 500. Calculate the percent error between the model and the actual data. Find all intercepts and any asymptotes for this function for $[S] \geq 0$. Which model would you say is better and why?

2. This problem is probably best done using Maple. Consider the quadratic function

$$f(x) = x^2 - 3x - 5$$

and the rational function

$$g(x) = \frac{20x}{1.4 + x}.$$

(You will probably want to graph these functions on the interval $x \in [-10, 10]$ with the range restricted to $y \in [-50, 50]$.)

a. Find the x and y-intercepts for both of these functions. Find the vertex of the quadratic function, $f(x)$. Give both x and y values. List any asymptotes (vertical and horizontal) for the rational function, $g(x)$.

b. Find all points of intersection between the graphs of $f(x)$ and $g(x)$.

[1] Schmider J., Greenblatt D.J., Harmatz J.S., Shader R.I., (1996) Enzyme kinetic modelling as a tool to analyse the behaviour of cytochrome P450 catalysed reactions: application to amitriptyline N-demethylation, *Brit. J. Clin. Pharmoco.* **41**, 593-604.