This assignment deals with fitting of the Michaelis Menten kinetics for a given enzyme. Substrate concentration [S] and the rate normalized to the enzyme concentration (i.e. $\mathrm{v} / \mathrm{E}_{0}$ ) is provided for your convenience. Use the approximation method that at high and low substrate concentration and obtain $\mathrm{k}_{\text {cat }}$ and $\mathrm{K}_{\mathrm{M}}$ from this procedure. Write the answer for the first 2 questions in the piece of paper, scan to make file 1. The following two questions carry 5 marks in total.

| $[\mathrm{S}]$ | $\mathrm{v} / \mathrm{E}_{0}$ |
| :---: | :---: |
| $(\mathrm{M})$ | $\left(\mathrm{s}^{-1} \cdot \mathrm{M}^{-1}\right)$ |
| 0 | 0.0000 |
| 0.0002 | 0.0020 |
| 0.0006 | 0.0055 |
| 0.001 | 0.0083 |
| 0.0025 | 0.0204 |
| 0.007 | 0.0436 |
| 0.01 | 0.0576 |
| 0.025 | 0.0871 |
| 0.06 | 0.1102 |
| 0.15 | 0.1347 |
| 0.25 | 0.1306 |
| 0.6 | 0.1371 |
| 1 | 0.1400 |
| 2 | 0.1449 |
| 5 | 0.1426 |

1. Use the data from [S] at 2.0 and 5.0 M to estimate $\mathrm{k}_{\text {cat. }}$ (round off to 3 decimals) Average of $k_{\text {cat }} 0.1449$ and 0.1426 is 0.1435 , which is rounded off to 0.144 .
2. Use the first 5 data points, use the $\mathrm{k}_{\text {cat }}$ from above and provide the value for $\mathrm{K}_{\mathrm{M}}$. Hint: set intercept to zero while fitting. (round off to 3 decimals)
Slope: 8.2353 units $=k_{\text {cat }} / K_{M}$
$\mathrm{K}_{\mathrm{M}}=\mathbf{0 . 0 1 7}$

[S]

Use the linearized form of the equation, i.e. the Lineweaver Burk plot, where you can plot $\mathrm{E}_{0} / v$ as a function of $1 /[\mathrm{S}]$. Save the matlab/excel sheet fits as a picture (file 2), using the numbers from this write the following in piece of paper and scan to make file 3 . The following two questions carry 10 marks in total.

3. Use the slope information to estimate $\mathrm{k}_{\text {cat }}$ (use the average value obtained from the fit only). (round off to 3 decimals)
Minor error in the question, intercept would provide $k_{\text {cat }}$
Intercept $=8.8622=1 / \mathrm{k}_{\text {cat }}$
$k_{\text {cat }}=0.113$ units
4. Use the intercept information to estimate $\mathrm{K}_{\mathrm{M}}$ (use the average value obtained from the fit only). (round off to 3 decimals)
Minor error in the question, slope with provide information of $\mathrm{K}_{\mathrm{M}}$
$K_{M}=0.0992^{*} 0.113=0.011$ units
Use the Michaelis Menten form to obtain the value of $\mathrm{k}_{\mathrm{cat}}$ and $\mathrm{K}_{\mathrm{M}}$. Fit using the cftool in MATLAB as taught in the lectures, save the final fit as a figure (file 4). The following question carries 15 marks in total. Write the following answers in a paper and scan it (file 5).

5. Write the average value of $\mathrm{k}_{\text {cat }}$ obtained from this method. (round off to 3 decimals) $k_{\text {cat }}=0.143$ units
6. Write the average value of $\mathrm{K}_{\mathrm{M}}$ obtained from this method. (round off to 3 decimals) $K_{M}=0.016$ units

