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.  $v/E_0$ ) is provided for your convenience.

Use the approximation method that at high and low substrate concentration and obtain  $k_{cat}$  and  $K_{M}$  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.

[S]	v/E <sub>0</sub>
(M)	(s <sup>-1</sup> .M <sup>-1</sup> )
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 k<sub>cat</sub>. (round off to 3 decimals)
- 2. Use the first 5 data points, use the  $k_{cat}$  from above and provide the value for  $K_{M\cdot}$  Hint: set intercept to zero while fitting. (round off to 3 decimals)

Use the linearized form of the equation, i.e. the Lineweaver Burk plot, where you can plot  $E_0/v$  as a function of 1/[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  $k_{cat}$  (use the average value obtained from the fit only). (round off to 3 decimals)
- 4. Use the intercept information to estimate  $K_M$  (use the average value obtained from the fit only). (round off to 3 decimals)

Use the Michaelis Menten form to obtain the value of  $k_{cat}$  and  $K_{M\cdot}$ . 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  $k_{\text{cat}}$  obtained from this method. (round off to 3 decimals)
- 6. Write the average value of  $K_{\text{M}}$  obtained from this method. (round off to 3 decimals)

Collate all the files (file 1 to file 5), make them into a single PDF file and upload.