Question 2: Enzyme Kinetics

1. Using the law of mass action, write down four equations for the rate of changes of the four species, *E*, *S*, *ES*, and *P*.

Solution:

The following equations can be obtained respectively according to the dynamic mass conservation for species *E*, *S*, *ES*, and *P*.

Rate of change of *E*:

$$\frac{d[E]}{dt} = -k_1[E][S] + k_3[ES] + k_2[ES]$$

Rate of change of *S*:

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

Rate of change of ES:

$$\frac{d[ES]}{dt} = k_1[E][S] - k_3[ES] - k_2[ES]$$

Rate of change of P:

$$\frac{d[P]}{dt} = k_3[ES]$$

where [*E*], [*S*], [*ES*], and [*P*] represent concentrations of species *E*, *S*, *ES*, and *P* respectively.

2. Write a code to numerically solve these four equations using the fourth-order Runge-Kutta method. For this exercise, assume that the initial concentration of *E* is 1 μM, the initial concentration of *S* is 10 μM, and the initial concentrations of *ES* and *P* are both 0. The rate constants are: *k1*=100/μM/min, *k2*=600/min, *k3*=150/min.

Solution:

The code written in Python to solve these four equations using the fourth-order Runge-Kutta method is as follows:

```
import math
import matplotlib.pyplot as plt
import numpy as np

def func_e(e,s,es,p):
    out = -100*e*s+150*es+600*es
    return out
def func_s(e,s,es,p):
    out = -100*e*s+600*es
    return out
def func_es(e,s,es,p):
    out = 100*e*s-150*es-600*es
```

```python
    return out
def func_p(e,s,es,p):
    out = 150*es
    return out

def runge_kutta_4(t0, e, s, es, p, h, tf):
    #t0: the initial time
    #e/s/es/p: the concentration of e/s/es/p
    #h: step length
    #tf: end time
    t = t0
    width = tf-t0+2
    length = 5
    data = {}
    while t <= tf:
        data[str(t)]=[]
        data[str(t)].append([e,s,es,p])

        K1 = func_e(e, s, es, p)
        L1 = func_s(e, s, es, p)
        M1 = func_es(e, s,es, p)
        N1 = func_p(e, s, es, p)

        K2 = func_e(e+K1*h/2, s+L1*h/2, es+M1*h/2, p+N1*h/2)
        L2 = func_s(e+K1*h/2, s+L1*h/2, es+M1*h/2, p+N1*h/2)
        M2 = func_es(e+K1*h/2, s+L1*h/2, es+M1*h/2, p+N1*h/2)
        N2 = func_p(e+K1*h/2, s+L1*h/2, es+M1*h/2, p+N1*h/2)

        K3 = func_e(e+K2*h/2, s+L2*h/2, es+M2*h/2, p+N2*h/2)
        L3 = func_s(e+K2*h/2, s+L2*h/2, es+M2*h/2, p+N2*h/2)
        M3 = func_es(e+K2*h/2, s+L2*h/2, es+M2*h/2, p+N2*h/2)
        N3 = func_p(e+K2*h/2, s+L2*h/2, es+M2*h/2, p+N2*h/2)

        K4 = func_e(e+K3*h, s+L3*h, es+M3*h, p+N3*h)
        L4 = func_s(e+K3*h, s+L3*h, es+M3*h, p+N3*h)
        M4 = func_es(e+K3*h, s+L3*h, es+M3*h, p+N3*h)
        N4 = func_p(e+K3*h, s+L3*h, es+M3*h, p+N3*h)

        e = e+(K1+2*K2+2*K3+K4) * h / 6
        s = s+(L1+2*L2+2*L3+L4) * h / 6
        es = es+(M1+2*M2+2*M3+M4) * h / 6
        p = p+(N1+2*N2+2*N3+N4) * h / 6

        t += h
```

```
    return data

def main():
    result = runge_kutta_4(0, 1, 10, 0, 0,0.00001, 1)

if __name__ == "__main__":
 main()
```

Where e, s, es, p in the code represents concentrations [*E*], [*S*], [*ES*], and [*P*] respectively, time step *h* is 0.00001 minute. Table 1 shows part of the numerical solution of the problem. Figure 1 shows the changes of concentration of *E, S*, *ES*, *P* with time according to the given initial conditions.

Table 1 Part of the numerical solution

t/min	0.00001	0.00002	0.00003	0.00004	0.00005	0.00006	0.00007	0.00008
[*E*]	9.90e-01	9.80e-01	9.71e-01	9.61e-01	9.52e-01	9.43e-01	9.34e-01	9.25e-01
[*S*]	9.99	9.98	9.97	9.96	9.95	9.94	9.93	9.93
[*ES*]	9.90e-03	1.96e-02	2.92e-02	3.86e-02	4.78e-02	5.68e-02	6.53e-02	7.44e-02
[*P*]	7.45e-06	2.96e-05	6.63e-05	1.17e-04	1.82e-04	2.60e-04	3.52e-04	4.57e-04

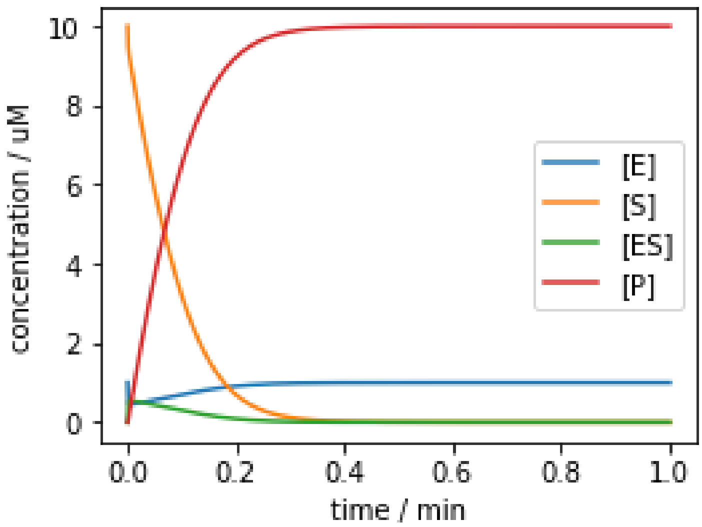

Figure 1 Changes of concentration of E, S, ES, P with time

3.We define the velocity, V, of the enzymatic reaction to be the rate of change of the product P. Plot the velocity V as a function of the concentration of the substrate S. You should find that, when the concentrations of S are small, the velocity V increases approximately linearly. At large concentrations of S, however, the velocity V saturates to a maximum value, Vm. Find this value Vm from your plot

Solution:

Figure 2 shows the change of enzymatic reaction velocity with concentration of the substrate S, th e maximum reaction velocity Vm is 141.10μM/min at 118.93μM of S.

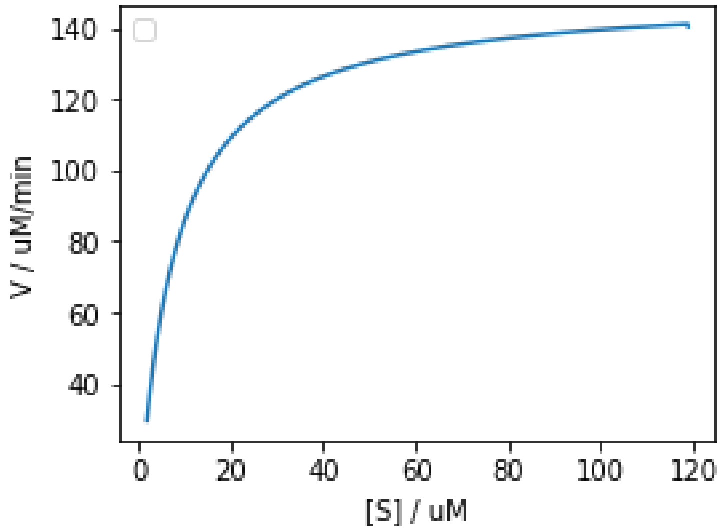

Figure 2 change of reaction velocity V with substrate *S* concentration