## **Enzyme Kinetics**

# **Questions and Examples**

3<sup>rd</sup> year undergraduates 2017-2018

**Q1** An enzyme hydrolyzed a substrate at 2 concentrations [S]= 0.03 and 0.12 mmol/L with Km value of around 0.06 mmol/L. The initial velocity observed at [S]= 0.03 mmol/L was  $1.5 \times 10^{-3}$  mmol/L.min-1. Calculate the initial velocity of the enzymatic reaction when using [S]= 0.12 mmol/L.

#### **Solution**

$$v_0 = \frac{V_{max}[S]}{K_m + [S]}$$

$$1.5*10^{-3} = \frac{\text{Vmax}* \ 0.03}{0.06 + 0.03}$$

Vmax = 
$$1.5*10^{-3}*3$$
  
= $4.5*10^{-3}$ 

$$V_o = \frac{4.5*10^{-3}*0.12}{0.06+0.12}$$

$$0.06 + 0.12$$
  $S1 = 0.03$   $S2 = 0.12$   $Km = 0.06$   $V_0 = 3*10^{-3}$   $V_{o1} = 1.5*10-3$ 

 $V_{02} = ?$ 

**Q2** An enzyme with a Km of 0.06 mmol/L hydrolyzed a substrate of a concentration 0.03 mmol/L. The initial velocity of the reaction was 0.0015 mmol/L.min <sup>-1</sup>. Calculate the substrate concentration which gives an initial velocity of 0.003 mmol/L.min<sup>-1</sup>.

#### **Solution**

$$v_0 = \frac{V_{max}[S]}{K_m + [S]}$$

$$1.5*10^{-3} = V_{\text{max}}*0.03$$
$$0.06 + 0.03$$

Vmax = 
$$1.5*10^{-3}*3$$
  
= $4.5*10^{-3}$ 

$$3*10^{-3} = 4.5*10^{-3} * S2$$
  
 $0.06 + S2$ 

S1= 0.03  
Km= 0.06  

$$V_{o1}$$
= 1.5\*10-3  
 $V_{02}$ = 3 \*10-3  
S2 =?

**Q3** An enzyme hydrolyzed a substrate concentration of 0.03 mmol/L, the initial velocity was  $1.5 \times 10^{-3}$  mmol/L.min<sup>-1</sup> and the maximum velocity was  $4.5 \times 10^{-3}$  mmol/L.min<sup>-1</sup>. Calculate the substrate concentration that gives a velocity of  $3 \times 10^{-3}$  mmol/L.min<sup>-1</sup>.

$$v_0 = \frac{V_{\text{max}}[S]}{K_{\text{m}} + [S]}$$

S1= 0.03  

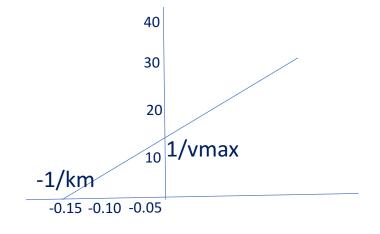
$$V_{o1}$$
= 1.5\*10-3  
 $V_{max}$ = 4.5\*10-3  
 $V_{o2}$ = 3 \*10-3  
S2 =?

**Q4** The following data describe an enzyme-catalysed reaction. Plot these results using the lineweaver-Burk method and determine values for Km and Vmax. The symbol mM represents m moles/L. The concentration of the enzyme is the same in all experiments.

[S] mM	Velocity mM/Sec
2.5	0.024
5	0.036
10	0.053
15	0.06
20	0.061

## **Solution**

1/[S] mM	1/Velocity mM/Sec
0.4	41.6
0.2	27.7
0.1	18.8
0.067	16.6
0.05	15.6



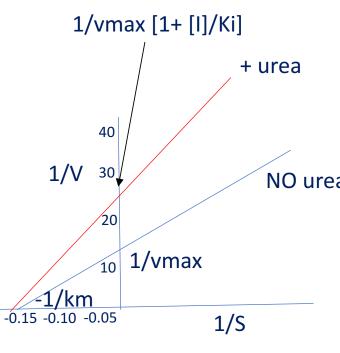
Q5 Sucrose is hydrolysed into glucose and fructose by the enzyme invertase. The reaction inhbited by addition of 2 M urea. Using the Lineweaver-Burk method of the following data to determine the type of the inhibition

Sucrose concentra tion Mol/L	Velocity mM/Sec	Velocity M/min + Urea (inhibitor)
0.02	0.18	0.08
0.05	0.26	0.11
0.08	0.31	0.15
0.11	0.33	0.16
0.17	0.37	0.19

## **Solution**

1/[S] mM	1/Velocity mM/Sec	1/V + I

Different slops, Different V max, same Km= non-competitive



**Q6** Alkaline phosphatase hydrolysed 2 mM of p-nitrophenol phosphate within 5 min. If the Vmax of the reaction was 4.5x10-3 mmol/L.min<sup>-1</sup> and the concentration of the product was 1.5\*10-3, how much do you expect the Km will be? when the reaction inhibited by 3 mM of Na-Pyrophoshate, the V max dropped to 1.5 \*10-3, what kind of inhibition was that?

**Q7** the hydrolysis of Phe-peptide is hydrolysed by chemotrypsin with the following result. Calculate Km and Vmax for the reaction

Peptide concentration M	Velocity mM/Sec
2.5*10-4	2.4*10-6
5*10-4	3.6*10-6
10*10-4	5.9*10-6
15*10-4	6*10-6

Q7 distinguish between the key-luck and the induced-fit models for binding E with S.

Q7 how can competitive and non-competitive inhibition be distinguished in terms of Km?