Problem 4-02 Constants of Michaelis-Menten equation from differential data; integrated equation ($c_{S0} << K_M$)

The values of initial reaction rates of enzyme reaction according to the above mentioned scheme for two different substrate concentrations are given in the following table:

c_S /(mmol dm^{-3})	0.25	76.923
$10^8\, \upsilon_0$ / (mol dm^{-3} s^{-1})	1.852	27.78

(a) Determine the constants of Michaelis-Menten equation K_M and υ_{max}.

(b) How long will it take to react 25 % of substrate, if its initial concentration was $c_{S0} = 2{\cdot}10^{-7}$ mol dm^{-3}?

[(a) $\upsilon_{max} = 2.9106 \cdot 10^{-7}$ mol dm^{-3} s^{-1}, $K_M = 3.679{\cdot}\ 10^{-3}$ mol dm^{-3}, (b) $\tau = 1.01$ h]

Solution:

$$r = -\frac{dc_S}{d\tau} = \frac{dc_P}{d\tau} = \frac{\upsilon_{max} \cdot c_S}{K_M + c_S} \qquad \text{kde} \quad \upsilon_{max} = k_2 \cdot c_{E0}$$

(a) Linearization (Lineweaver and Burke):

$$\frac{1}{\upsilon} = \frac{1}{\upsilon_{max}} + \frac{K_M}{\upsilon_{max}} \cdot \frac{1}{c_S}$$

$$\frac{1}{1.852 \cdot 10^{-8}} = \frac{1}{\upsilon_{max}} + \frac{K_M}{\upsilon_{max}} \cdot \frac{1}{0.25 \cdot 10^{-3}} \qquad (0.25 \text{ mmol dm}^{-3} = 0.25{\cdot}10^{-3} \text{ mol dm}^{-3})$$

$$\frac{1}{27.78 \cdot 10^{-8}} = \frac{1}{\upsilon_{max}} + \frac{K_M}{\upsilon_{max}} \cdot \frac{1}{76.923 \cdot 10^{-3}}$$

$$\frac{K_M}{\upsilon_{max}} = \frac{\dfrac{1}{1.852 \cdot 10^{-8}} - \dfrac{1}{27.78 \cdot 10^{-8}}}{\dfrac{1}{0.25 \cdot 10^{-3}} - \dfrac{1}{76.923 \cdot 10^{-3}}} = 12640 \text{ s}$$

$$\frac{1}{\upsilon_{max}} = \frac{1}{1.852 \cdot 10^{-8}} - 12640 \cdot \frac{1}{0.25 \cdot 10^{-3}} = 3435680.346 \quad , \upsilon_{max} = 2.9106 \cdot 10^{-7} \text{ mol dm}^{-3} \text{ s}^{-1}$$

$K_M = 12640 \cdot 2.9106 \cdot 10^{-7} = 3.679{\cdot}\ 10^{-3}$ mol dm^{-3}

(b) $c_{S0} = 2{\cdot}10^{-7}$ mol dm^{-3} << $K_M = 3.679{\cdot}\ 10^{-3}$ mol dm^{-3}

$$-\frac{dc_S}{d\tau} = \frac{\upsilon_{max} \cdot c_S}{K_M} \quad \text{- first-order reaction}$$

balance: $c_S = c_{S0} - c_{S0} \cdot \alpha \quad , \quad d\,c_S = -\,c_{S0} \cdot d\alpha$

$$+\frac{c_{S0}\, d\alpha}{d\tau} = \frac{\upsilon_{max}}{K_M} \cdot c_{S0} \cdot (1-\alpha)$$

$$-\ln(1-\alpha) = \frac{\upsilon_{max}}{K_M} \cdot \tau$$

$$\tau = -\frac{K_M}{\upsilon_{max}} \cdot \ln(1-\alpha) = -\frac{3.679 \cdot 10^{-3}}{2.9106 \cdot 10^{-7}} \cdot \ln(1-0.25) = 3636.3 \text{ s} = 1.01 \text{ h}$$