Problem 4-09 Integrated Michaelis-Menten equation; $c_{S0} << K_M$; molar enzyme activity

Michaelis constant K_M = 226 mmol dm^{-3} was evaluated from the measurements of initial reaction rates for the enzymatic reaction described by the following scheme:

$$\mathrm{E + S} \underset{k_{-1}}{\overset{k_{+1}}{\rightleftarrows}} \mathrm{ES} \xrightarrow{k_2} \mathrm{E + P_1 + P_2}$$

At enzyme concentration 2.17·10^{-6} mol dm^{-3} the concentration of substrate (M_S = 150 g mol^{-1}) decreased to 40 % of the initial value 3 μg cm^{-3} in 1.2 hour. Calculate the molar activity of the enzyme catalyzing the above mentioned reaction. [k_2 = 22.09 s^{-1}]

Solution:

w_{S0} = 3 μg cm^{-3} = 3·10^{-6} g (10^3 dm^{-3}) = 3·10^{-3} g dm^{-3}
M_S = 150 g mol^{-1}

$$\left.\begin{aligned} c_{S0} &= \frac{w_{S0}}{M_S} = \frac{3\cdot 10^{-3}}{150} = 2\cdot 10^{-5}\ \mathrm{mol\ dm^{-3}} \\ K_M &= 226\ \mathrm{mmol\ dm^{-3}} = 0.226\ \mathrm{mol\ dm^{-3}} \end{aligned}\right\} \qquad c_{S0} << K_M$$

τ = 1.2 h = 4320 s
c_S = 0.4 c_{S0}
c_{E0} = 2.17·10^{-6} mol dm^{-3}

$$\upsilon = \frac{\mathrm{d}c_P}{\mathrm{d}\tau} = -\frac{\mathrm{d}c_S}{\mathrm{d}\tau} = \frac{\upsilon_{max}\cdot c_S}{K_M + c_S} \cong \frac{\upsilon_{max}}{K_M}\cdot c_S$$

- first-order kinetics with the rate constant = υ_{max}/K_M

$$-\int_{c_{S0}}^{c_S} \frac{\mathrm{d}c_S}{c_S} = \frac{\upsilon_{max}}{K_M}\cdot\int_0^{\tau} \mathrm{d}\tau$$

$$-\ln\frac{c_S}{c_{S0}} = \frac{\upsilon_{max}}{K_M}\cdot\tau \quad , \quad \upsilon_{max} = k_2\cdot c_{E0}$$

$$k_2 = \frac{K_M}{c_{E0}\cdot\tau}\cdot\ln\frac{c_{S0}}{c_S} = \frac{0.226}{2.17\cdot 10^{-6}\cdot 4320}\cdot\ln\frac{c_{S0}}{0.4\,c_{S0}} = 22.09\ \mathrm{s^{-1}}$$