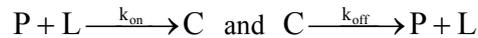


Problem 1.

The binding of protein P with ligand L to form complex C is reversible, as told



We are given a table with various initial concentrations of L in order to estimate k_{on} and k_{off} and also K_d for the reaction.

$$\frac{dC_C}{dt} = k_{on} C_P C_L - k_{off} C_C$$

Also from material balances and stoichiometry, we have $C_P + C_C = C_{P0}$ and $C_L + C_C = C_{L0}$, therefore

$$\frac{dC_C}{dt} = k_{on} (C_{P0} - C_C)(C_{L0} - C_C) - k_{off} C_C$$

In this problem, we can safely assume that $C_{L0} - C_C \approx C_{L0}$ since $C_{L0} \gg C_{P0}$ in all three cases of different C_{L0} .

Thus, the integrated analytic expression for C_C becomes

$$C_C = \frac{k_{on} C_{P0} C_{L0}}{k_{on} C_{L0} + k_{off}} \{1 - \exp[-(k_{on} C_{L0} + k_{off})t]\} = \frac{C_{P0} C_{L0}}{C_{L0} + K_d} \{1 - \exp[-(k_{on} C_{L0} + k_{off})t]\}$$

where $K_d \equiv \frac{k_{off}}{k_{on}}$

Therefore, if we plot C_C w.r.t time for each cases of C_{L0} , we can fit according to an exponential

$y = a[1 - \exp(-bt)]$, where b is $k_{on} C_{L0} + k_{off}$, and a is $\frac{C_{P0} C_{L0}}{C_{L0} + K_d}$. Values for a and b are shown in the

following table.

L_0 (μ M)	a	b
1	0.903	1.1113
5	1.0436	4.7047
15	0.9932	15.1079

One important observation in this table is that parameter a does not change much when initial ligand concentration is changed, indicating $C_{L0} = 1 \mu\text{M}$ is already above the saturating value. Therefore, value for k_{off} can not be obtained accurately from this design of experiments. We can only conclusively obtain the value for k_{on} .

So we fit a linear express of b vs. C_{L0} to get k_{on} .

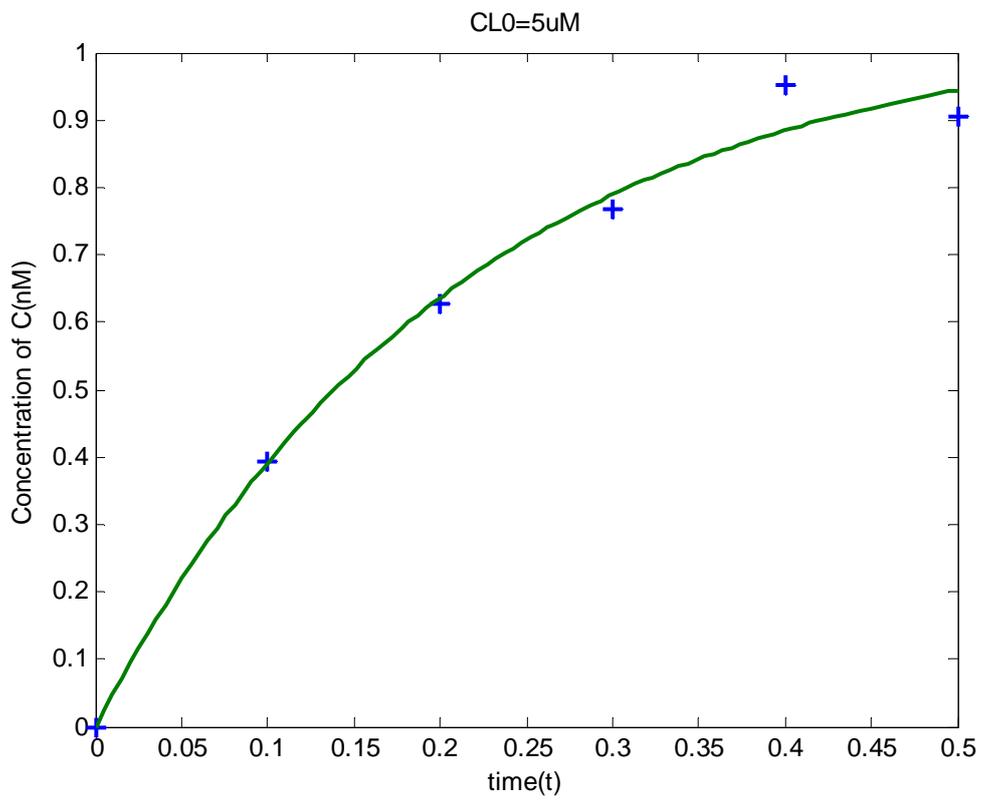
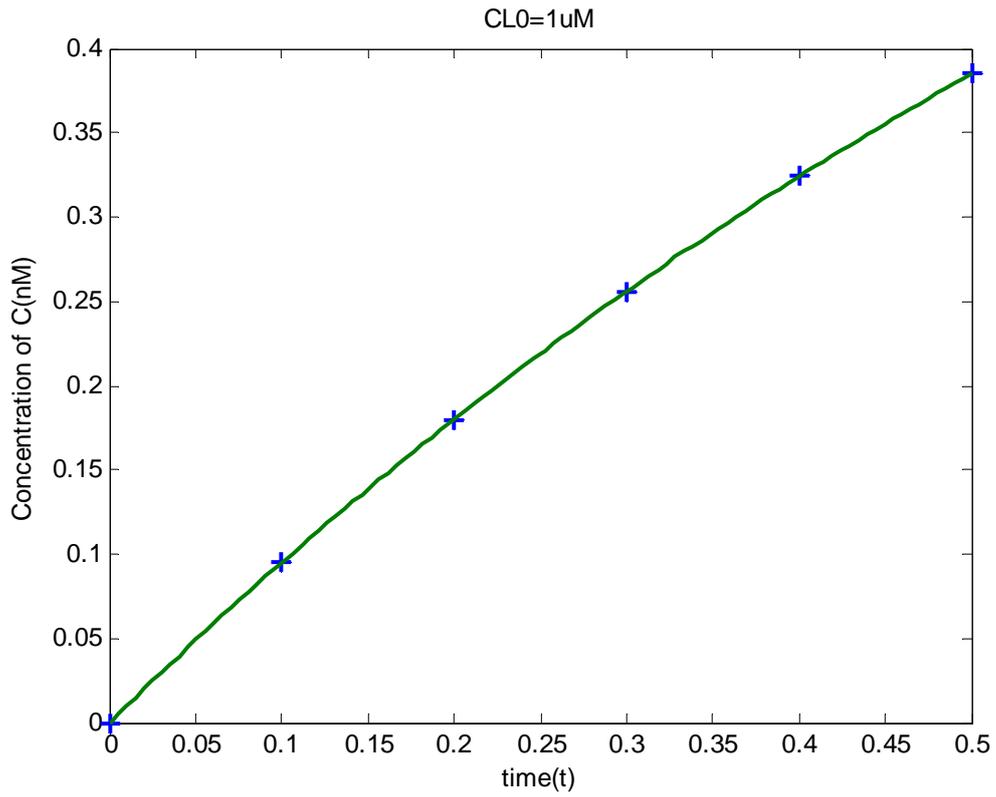
$$k_{on} = 0.0010 \text{ nM}^{-1} \text{ sec}^{-1}$$

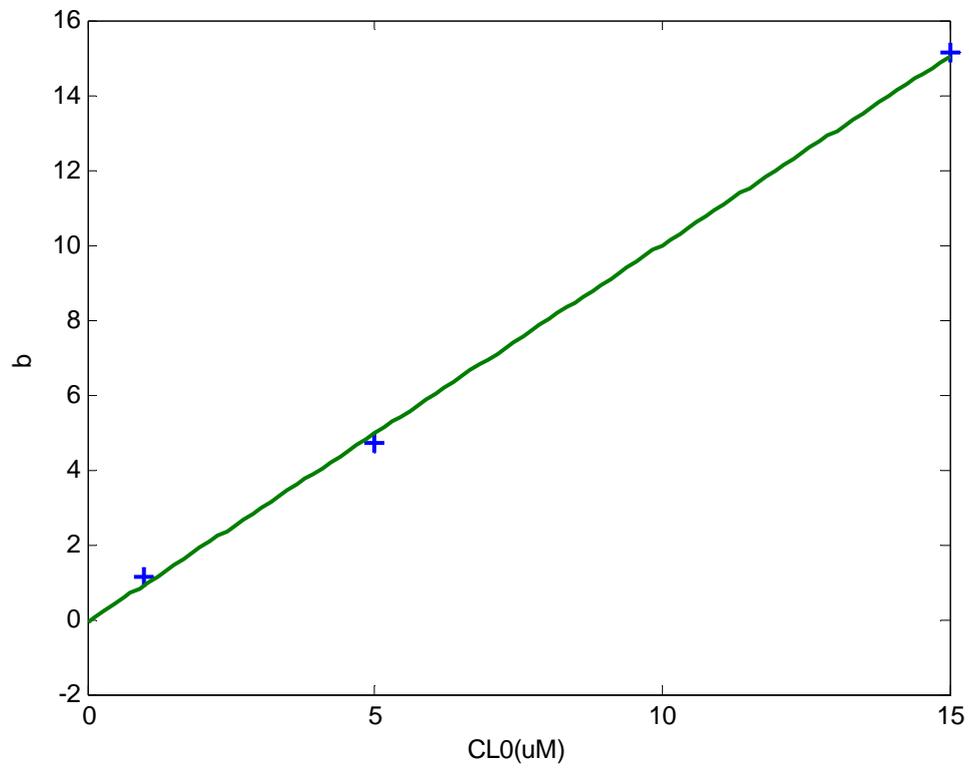
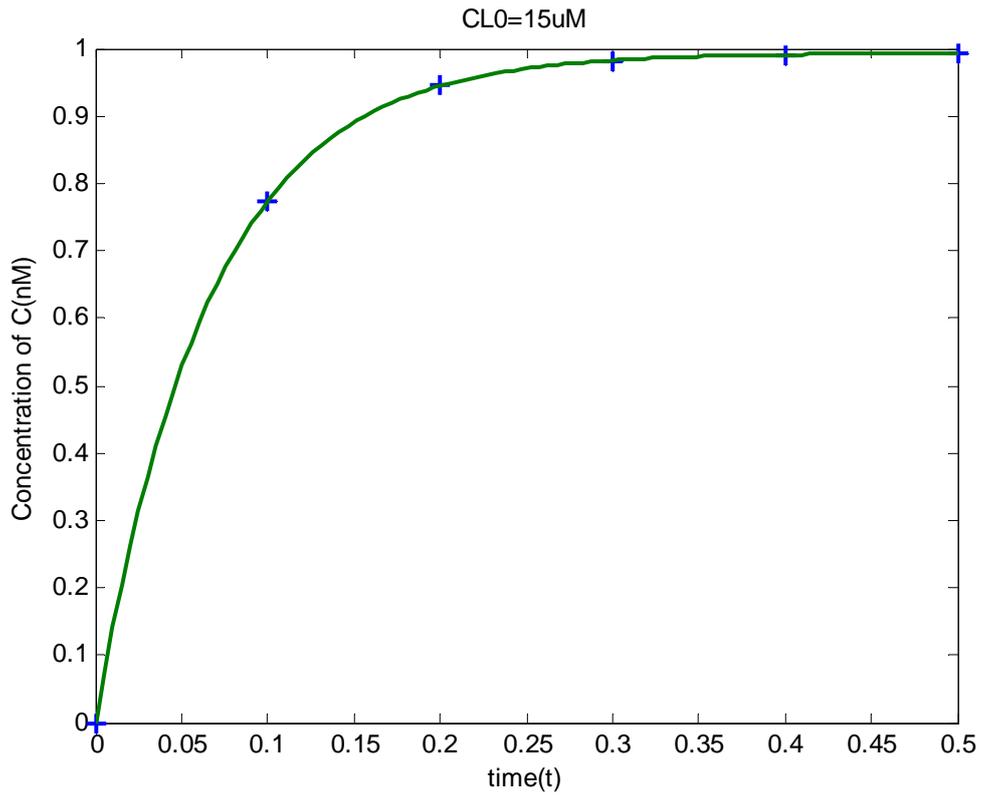
An estimate on k_{off} would be

$$0 \leq k_{off} \ll k_{on} C_{L0, \min} = 0.0010 \text{ nM}^{-1} \text{ sec}^{-1} * 1 \mu\text{M} = 1 \text{ sec}^{-1}$$

Similarly an estimate on K_d is

$$0 \leq K_d \ll C_{L0, \min} = 1 \mu\text{M}.$$





Problem 2.

For a steady state chemostat, the material balance on cell mass yields

$$D \equiv \frac{F}{V} = \mu$$

the volumetric productivity is

$$F(X - X_0) = \mu V(X - X_0)$$

For a batch reactor, the material balance on cell mass yields

$$\frac{dX}{dt} = \mu X$$

where the initial condition is $X(t=0) = X_0$.

Therefore, we have $X = X_0 \exp(\mu t)$

The volumetric productivity is

$$\frac{V(X - X_0)}{t + t_{\text{turn}}} = \frac{V(X - X_0)}{\frac{1}{\mu} \ln \frac{X}{X_0} + t_{\text{turn}}}$$

Therefore, the ratio of the two

$$\frac{\mu V(X - X_0)}{\frac{V(X - X_0)}{\frac{1}{\mu} \ln \frac{X}{X_0} + t_{\text{turn}}}} = \ln \frac{X}{X_0} + \mu t_{\text{turn}}$$

In practice, for chemostat, in order to maximize the productivity of biomass (DX), the operating condition for μ is close μ_{max} . Therefore the ratio above is approximately

$$\ln \frac{X}{X_0} + \mu_{\text{max}} t_{\text{turn}}$$

Problem 3.

The expression that would be suitable to describe the change of protein expression is:

$$C_p = \frac{k_p k_r}{\gamma_r (\gamma_p + \mu)} \{1 - \exp[-(\gamma_p + \mu)t]\}$$

where the meaning of each symbol is in accord with what we did in class.

The time required to change from an “off state” to an “on state” (95% of the steady-state value) is

$$0.95 = 1 - \exp[-(\gamma_p + \mu)t] \quad \text{or} \quad t = -\frac{\ln 0.05}{\gamma_p + \mu}$$

- a) if cells are rapidly growing with a doubling time 30 min and stable protein with a degradation half-time one day, i.e.

$$\frac{\ln 2}{\mu} = 30 \text{ min} \quad \text{and} \quad \frac{\ln 2}{\gamma_p} = 1 \text{ day}$$

So the half-time for switching is the time need to reach

$$0.95 \times \frac{1}{2} = 1 - \exp[-(\gamma_p + \mu)t]$$

$$t_{\text{switching}} = \frac{-\ln(1 - 0.95 \times \frac{1}{2})}{(\gamma_p + \mu)} = \frac{-\ln(1 - 0.95 \times \frac{1}{2})}{\frac{\ln 2}{30 \text{ min}} + \frac{\ln 2}{1 \text{ day}}} \approx 27.9 \text{ min}$$

- b) if cells are not growing at all and the protein with a degradation half-time one hour, i.e.

$$\frac{\ln 2}{\gamma_p} = 1 \text{ hr}$$

So the half-time for switching is

$$t_{\text{switching}} = \frac{-\ln(1 - 0.95 \times \frac{1}{2})}{(\gamma_p + \mu)} = \frac{-\ln(1 - 0.95 \times \frac{1}{2})}{\frac{\ln 2}{1 \text{ hr}} + 0} = 0.93 \text{ hr}$$

So in both cases, the switching times are much much longer than that of the current electronic circuits, which is on the order of ~ns-μs. Thus, it would not be promising in realizing a computer for practical uses.