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Prelim 1

1.a)

$$R_{x,T} = R_x^0 + (G_j : R_x)_c + (G_j : R_x)_0 + \sum_{i=1,j}^{N} \left[(G_i : R_x)_c + (G_i : R_x)_0 \right]$$

From the proposed reactions, the open and close complexes balances are as follows:

$$\frac{d}{dt} (G_j : R_x)_c = (k_{+,j} \, G_j \, R_x^0) - k_{-,j} (G_j : R_x)_c - k_{I,j} (G_j : R_x)_c$$

$$\frac{d}{dt} (G_j : R_x)_0 = k_{I,j} (G_j : R_x)_c - k_{A,j} (G_j : R_x)_0 - k_{E,j} (G_j : R_x)_0$$

At steady-state:

① $(G_j : R_x)_c = \dfrac{k_{+,j} \, G_j \, R_x^0}{k_{-,j} + k_{I,j}}$ → If we define $\quad k_{x,j} = \dfrac{k_{-,j} + k_{I,j}}{k_{+,j}}$

$\quad (G_j : R_x)_c = \dfrac{G_j \, R_x^0}{k_{x,j}}$

② $(G_j : R_x)_0 = \dfrac{k_{I,j} (G_j : R_x)_c}{k_{A,j} + k_{E,j}}$ → If we define $\quad \tau_{x,j} = \dfrac{k_{A,j} + k_{E,j}}{k_{I,j}}$

$\quad (G_j : R_x)_0 = \dfrac{(G_j : R_x)_c}{\tau_{x,j}}$

By replacing ① in ② :

③ $(G_j : R_x)_0 = \dfrac{G_j \, R_x^0}{k_{x,j} \, \tau_{x,j}}$ → where R_x^0 is the amount of free RNAP.

Note that the same is true for any gene (G_i)

At steady state, the amount of R_x^0 is given by:

$$R_x^0 = R_{x,T} - (G_j:Rx)_c - (G_j:Rx)_0 - \sum_{i=1,j}^{N} \left[(G_i:Rx)_c + (G_i:Rx)_0 \right]$$

$$R_x^0 = R_{x,T} - \frac{G_j R_x^0}{K_{x,j}} - \frac{G_j R_x^0}{K_{x,j} T_{x,j}} - \sum_{i=1,j}^{N} \left[\frac{G_i R_x^0}{K_{x,i}} + \frac{G_i R_x^0}{K_{x,i} T_{x,i}} \right]$$

$$K_{x,j} T_{x,j} R_x^0 = K_{x,j} T_{x,j} R_{x,T} - G_j R_x^0 T_{x,j} - G_j R_x^0 - \sum_{i=1,j}^{N} \left[\frac{G_i R_x^0 K_{x,j} T_{x,j}}{K_{x,i} T_{x,i}} (T_{x,i} + 1) \right]$$

$$K_{x,j} T_{x,j} R_{x,T} = R_x^0 \left[K_{x,j} T_{x,j} - G_j T_{x,j} - G_j - \sum_{i=1,j}^{N} \frac{K_{x,j} T_{x,j}}{K_{x,i} T_{x,i}} (1 + T_{x,i}) G_i \right]$$

$\hookrightarrow$ Let $\varepsilon_j = \sum_{i=j}^{N} \frac{K_{x,j} T_{x,j}}{K_{x,i} T_{x,i}} (1 + T_{x,i}) G_i$

$$K_{x,j} T_{x,j} R_{x,T} = R_x^0 \left(K_{x,j} T_{x,j} - G_j T_{x,j} - G_j - \varepsilon_j \right)$$

④ $\quad R_x^0 = \dfrac{K_{x,j} T_{x,j} R_{x,T}}{K_{x,j} T_{x,j} - G_j T_{x,j} - G_j - \varepsilon_j}$

Replacing ④ in ③ =

$$(G_j:Rx)_0 = R_{x,T} \left(\frac{G_j}{K_{x,j} T_{x,j} + (1 + T_{x,j}) G_j + \varepsilon_j} \right)$$

$\hookrightarrow$ which leads to

$$r_{x,j} = k_{\varepsilon_j} R_{x,T} \left(\frac{G_j}{K_{x,j} T_{x,j} + (1 + T_{x,j}) G_j + \varepsilon_j} \right) \quad \square$$

b) For the N-gene system to be equal to the 1-gene system, $\varepsilon_j \ll K_{x,j} T_{x,j}$ and $\varepsilon_j \ll (1 + T_{x,j}) G_j$. Without getting into the numbers needed for that to be true, one can think in terms of time scales. As long as the transcription of gene j is much more slower or much more faster than that of all other N-1 genes, the system will behave as a 1-gene system.

2.

$$\text{Inducer} \longrightarrow P_1 \longrightarrow P_2 \longrightarrow P_3$$
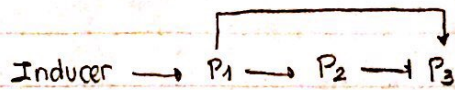
$$\frac{dm_1}{dt} = r_{x_1} u(I) - m_1(D_{x_1} + \mu)$$

$$\frac{dm_2}{dt} = r_{x_2} u(P_1) - m_2(D_{x_2} + \mu)$$

$$\frac{dm_3}{dt} = r_{x_3} u(P_1, P_2) - m_3(D_{x_3} + \mu)$$

$$\frac{dP_1}{dt} = r_{L_1} \omega(m_1) - P_1(D_{L_1} + \mu)$$

$$\frac{dP_2}{dt} = r_{L_2} \omega(m_2) - P_2(D_{L_2} + \mu)$$

$$\frac{dP_3}{dt} = r_{L_3} \omega(m_3) - P_3(D_{L_3} + \mu)$$

$$\frac{dx_j}{dt} = Sr + Ax$$

$$S = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 & 0 \\ 0 & 1 & 0 & 0 & 0 & 0 \\ 0 & 0 & 1 & 0 & 0 & 0 \\ 0 & 0 & 0 & 1 & 0 & 0 \\ 0 & 0 & 0 & 0 & 1 & 0 \\ 0 & 0 & 0 & 0 & 0 & 1 \end{bmatrix} \qquad \Gamma = \begin{bmatrix} r_{x_1} u(I) \\ r_{x_2} u(P_1) \\ r_{x_3} u(P_1, P_2) \\ r_{L_1} \omega(m_1) \\ r_{L_2} \omega(m_2) \\ r_{L_3} \omega(m_3) \end{bmatrix}$$

$$A = \begin{bmatrix} -(D_{x_1}+\mu) & 0 & 0 & 0 & 0 & 0 \\ 0 & -(D_{x_2}+\mu) & 0 & 0 & 0 & 0 \\ 0 & 0 & -(D_{x_3}+\mu) & 0 & 0 & 0 \\ 0 & 0 & 0 & -(D_{L_1}+\mu) & 0 & 0 \\ 0 & 0 & 0 & 0 & -(D_{L_2}+\mu) & 0 \\ 0 & 0 & 0 & 0 & 0 & -(D_{L_3}+\mu) \end{bmatrix}$$

$$x = \begin{bmatrix} m_1 \\ m_2 \\ m_3 \\ P_1 \\ P_2 \\ P_3 \end{bmatrix}$$

where the control functions are given by:

$$u(I) = \frac{W_{11} + W_{I1}\, f_{I1}}{1 + W_{11} + W_{I1}\, f_{I1}}$$

$$u(P_1) = \frac{W_{22} + W_{12}\, f_{12}}{1 + W_{22} + W_{12} f_{12}}$$

$$u(P_1, P_2) = \frac{W_{33} + W_{13} f_{13}}{1 + W_{33} + W_{13} f_{13} + W_{23} f_{23}}$$

$$f_{I1} = \frac{I^n}{k_I^n + I^n} \qquad f_{12} = \frac{P_1^n}{k_{12}^n + P_1^n} \qquad f_{13} = \frac{P_1^n}{k_{13}^n + P_1^n} \qquad f_{23} = \frac{P_2^n}{k_{23}^n + P_2^n}$$

Parameters:

$k_I = 0.3$	$n_I = 1.5$	$W_{I1} = 100$
$k_{12} = 1.0$	$n_{12} = 1.5$	$W_{11} = W_{22} = W_{33} = 0.000001$
$k_{13} = 1.0$	$n_{13} = 1.5$	$W_{12} = 10.0$
$k_{23} = 1.0$	$n_{23} = 10$	$W_{13} = 5.0$
		$W_{23} = 25.0$

$T_d = 40$ min

$k_L = 454.64$ nmol /gDW

$r_x = 1150$ copies/cell $\rightarrow$ RNAP

$r_L = 45000$ copies/cell $\rightarrow$ Ribosomes

$C_p = 200$ copies/cell

$L_x = 1000$ nt

$L_T = 333$ AA

$d_{x1} = 1200$ nt

$d_{x2} = 2400$ nt

$d_{x3} = 600$ nt

$d_{L1} = 400$ AA

$d_{L2} = 800$ AA

$d_{L3} = 200$ AA

$C_W = 2.8 \times 10^{-13}$ g/cell

$H_2O\,W = 0.7$

$t_{1/2} = 2.1$ min $\rightarrow$ mRNA

$t_{1/2} = 24$ h $\rightarrow$ protein

$\dot{v}_x = 60$ nts/s $\rightarrow$ elongation rate (RNAP)

$\dot{v}_L = 16.5$ aa/s $\rightarrow$ elongation rate (ribosomes)

$k_x = 0.24$ nmol /gDW

2.C. From singular value decomposition:

• Steady-State ranking:

1. P_3
2. P_1
3. P_2
4. mRNA 3
5. mRNA 1
6. mRNA 2

• Early-inducer ranking:

1. P_1
2. P_2
3. P_3
4. mRNA 1
5. mRNA 2
6. mRNA 3

• Late-inducer ranking:

1. P_1
2. P_2
3. P_3
4. mRNA 1
5. mRNA 2
6. mRNA 3

Singular value decomposition allows to rank the sensitivity of species to changes in the basic parameters. During steady-state without inducer, P_3 is the most sensitive specie. This shifts to P_1 after the inducer is introduced. Absolute values of the first column of the U matrix for each time period are presented in matrices U_1, U_2 and U_3, respectively. The shift in rankings after the inducer is introduced is expected due to the weight of the inducer in the expression of P_1.

3. FBA

$d_U = 308$ aa

$d_{X1} = 924$ nt

$G_P = 5$nM $\rightarrow$ Plasmid concentration

$C_V = 15\,\mu L$

bounds $= \pm\ 100\,000\ \mu M/h.$

$R_X = 0.15\ \mu M \longrightarrow$ RNAP

$R_L = 1.6\ \mu M \longrightarrow$ ribosomes

$\dot{v}_X = 60$ nts/s

$\dot{v}_L = 16.5$ aa/s

$K_X = 0.3\mu M$

$K_L = 57\ \mu M$

$\tau_X = 2.7$

$\tau_L = 0.8$

$K_{dx} = 8.35\ h^{-1}$

$K_{dL} = 9.9 \times 10^{-3}\ h^{-1}$

$d_X = 1000$ nt

$d_L = 330$ aa.

3.a.

Reactions:

$$G + RNAP \xrightarrow{v_1} G^*$$

$$G^* + nNTP \xrightarrow{v_2} mRNA + G + RNAP + 2nP_i$$

$$mRNA + rib \xrightarrow{v_4} rib^*$$

$$mRNA \xrightarrow{v_3} nNMP$$

$$rib^* + aAAtRNA + 2aGTP \xrightarrow{v_5} atRNA + 2aGDP + 2aPi + rib + mRNA + protein$$

$$AA + tRNA + ATP \xrightarrow{v_6} AMP + 2Pi + AAtRNA$$

Source $\xrightarrow{b_1}$ AA   Protein $\xrightarrow{b_3}$ sink

Source $\xrightarrow{b_2}$ NTP   NMP $\xrightarrow{b_4}$ sink

Source $\xrightarrow{b_5}$ ATP   AMP $\xrightarrow{b_6}$ sink

Source $\xrightarrow{b_7}$ GTP   GDP $\xrightarrow{b_8}$ sink

Pi $\xrightarrow{b_9}$ sink
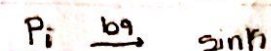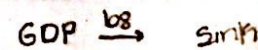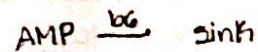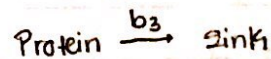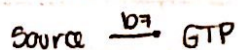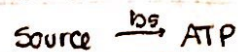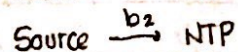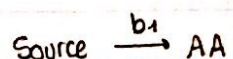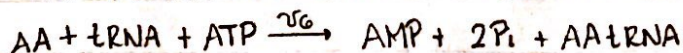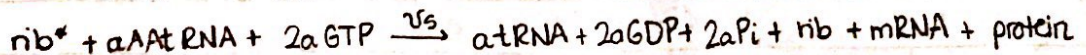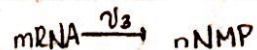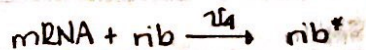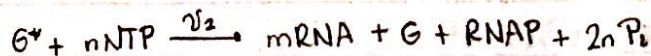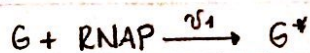
Stochiometric matrix:

	v_1	v_2	v_3	v_4	v_5	v_6	b_1	b_2	b_3	b_4	b_5	b_6	b_7	b_8	b_9
G	-1	1	0	0	0	0	0	0	0	0	0	0	0	0	0
RNAP	-1	1	0	0	0	0	0	0	0	0	0	0	0	0	0
G*	1	-1	0	0	0	0	0	0	0	0	0	0	0	0	0
NTP	0	-n	0	0	0	0	0	1	0	0	0	0	0	0	0
mRNA	0	1	-1	-1	1	0	0	0	0	0	0	0	0	0	0
Pi	0	2n	0	0	2a	2	0	0	0	0	0	0	0	0	-1
NMP	0	0	n	0	0	0	0	0	0	-1	0	0	0	0	0
rib	0	0	0	-1	1	0	0	0	0	0	0	0	0	0	0
rib*	0	0	0	1	-1	0	0	0	0	0	0	0	0	0	0
AAtRNA	0	0	0	0	-a	1	0	0	0	0	0	0	0	0	0
GTP	0	0	0	0	-2a	0	0	0	0	0	0	0	1	0	0
tRNA	0	0	0	0	a	-1	0	0	0	0	0	0	0	0	0
GDP	0	0	0	0	2a	0	0	0	0	0	0	0	0	-1	0
Protein	0	0	0	0	1	0	0	0	-1	0	0	0	0	0	0
AA	0	0	0	0	0	-1	1	0	0	0	0	0	0	0	0
ATP	0	0	0	0	0	-1	0	0	0	0	1	0	0	0	0
AMP	0	0	0	0	0	1	0	0	0	0	0	-1	0	0	0

$\llcorner$ where $v_2 = \hat{r}_X$ and $0 \le v_5 \le \hat{r}_L$

• From the models in class, let $\hat{r}_X$ and $\hat{r}_L$ be given by:

$$\hat{r}_X = \dot{v}_X \, R_X \left(\frac{G_p}{K_X \tau_X + (1 + \tau_X) G_p} \right) \qquad ①$$

$$\hat{r}_L = \dot{v}_L \, R_L \left(\frac{mRNA}{K_L \tau_L + (1 + \tau_L) mRNA} \right) \qquad ②$$

• Bounds for all other rates will be set to $0 \le v_x \le 100\,000$ and $-100\,000 \le b_x \le 100\,000$